The University of Chicago

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A DISSERTATION SUBMITTED TO THE FACULTY
OF THE BIOLOGICAL SCIENCES IN CANDIDACY
FOR THE DEGREE OF DOCTOR OF PHILOSOPHY

DEPARTMENT OF BOTANY

By
ALICE ALLEN BAILEY

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EFFECTS OF ULTRAVIOLET RADIATION UPON REPRESENTATIVE SPECIES OF FUSARIUM[1]

ALICE ALLEN BAILEY

(WITH PLATE I AND SIX FIGURES)

Introduction

One of the most common fungi with which the market pathologist must deal is the ubiquitous *Fusarium*. There are very few vegetables on which *Fusarium* does not occur, either as a primary or secondary incitant of decay, and so the problem of identification of *Fusarium* species is an ever present one. During the course of recent work with species of *Fusarium* isolated from decayed onion bulbs, one of the chief difficulties has been the production in artificial culture of sufficient normal macrospores for taxonomic studies. Various methods of inducing sporulation have been tried, but in many cases these were unsuccessful or only inconsistently fruitful. Often microspores were produced in abundance but the macrospores, which are usually necessary for identification, were lacking. The consistent success of RAMSEY and BAILEY (14) in producing macrospores of the onion parasite *Fusarium cepae* Hanz. emend. LINK and BAILEY (8) upon irradiation with ultraviolet suggested the use of this method for unidentified strains of *Fusarium* from onion. In not all instances was the irradiation as beneficial as in the case of *F. cepae*, but in a large percentage of strains it was helpful and in some it induced marked increase in spore production.

[1] Contribution from the Division of Horticultural Crops and Diseases, Bureau of Plant Industry, United States Department of Agriculture and the Department of Botany, University of Chicago, cooperating.

Because of the value of ultraviolet radiation in the work on the taxonomy of onion Fusaria, irradiation of authentic species of *Fusarium* from the various sections of the genus was tried, in the hope that this might prove helpful in the ever difficult problem of taxonomic diagnosis of the many species within the form genus. Investigations on the effect of ultraviolet irradiation upon members of the genus *Fusarium* have been made by others, but most of the work has dealt with the lethal action of the rays. SIBILIA (18) describes the lethal effect of ultraviolet on two species of *Fusarium*, *F. echinosporum* and *F. fuliginosporum*, first described by him (17) but not mentioned by WOLLENWEBER in the Fusaria Autographice Delineata (29, 30). SIBILIA found that exposure of the conidia of these two strains in drops of distilled water or of 1 per cent glucose solution at a distance of 12 cm. from the unscreened quartz mercury vapor lamp for two minutes or over invariably resulted in death. The age of the conidia seemed to play no part in their resistance to ultraviolet. Chlamydospores were more resistant than conidia. In most of the experiments, exposures as short as one second, even at 90 cm. from the arc, resulted in death; however a distance of 2 meters permitted germination of the spores, the air screening out the more detrimental rays. The contents of spores which were killed by radiations were greatly plasmolyzed, and showed numerous small globular masses or, rarely, a single ellipsoidal mass. Conidia germinating under irradiation had abnormal germ tubes, often contorted and soon arrested in growth.

FULTON and COBLENTZ (5), working with *Fusarium* sp. from orange fruit, found that the spores, in water suspensions, were killed by 1-minute exposures 6 inches from an unscreened 110-volt quartz mercury burner having a mercury cathode and a tungsten anode, operated on 320 watts (80 volts, 4 amperes). Mycelium from germinated spores was more easily killed than were ungerminated spores of the various fungi tested. The fungicidal effectiveness of treatment with ultraviolet depended on the intensity of the radiation and on the wave lengths of the component rays; the shorter the wave length the more lethal the effect with a given intensity. The killing effect was cumulative, several short intermittent exposures (rest periods were the same length as exposure periods) having almost the

effect of one continuous exposure of the same total time. The killing effect increased with the lengthening of the time of action, and also with the increase in intensity of irradiation.

DILLON WESTON and HALNAN (4) also worked on *Fusarium* sp. and found that the mycelium was not killed but that growth was inhibited. Their tests were not run with an unscreened lamp, however, but with vitaglass and Sanalux petri dish covers transmitting only 50 to 70 per cent of the ultraviolet light from a quartz mercury-vapor lamp (Hanovia artificial alpine Sun, 2.5 amp. 210 v. direct current type). Potato agar plates were inoculated from a pure culture of the *Fusarium*, a needle being used to carry the spores and mycelium into the medium. Irradiation was commenced the same day and was continued for a fortnight with daily 9-minute exposures. They found that the mycelium of *Mucor* and *Fusarium* grew farther down into the medium in the irradiated plates; "consequently, aerial development is retarded and the sporangiophores and conidiophores appear stunted." After the first week of irradiation the plates were reversed and the unirradiated controls were subjected to a week's treatment while the previously treated plates were held as controls. "The effect of this treatment was to inhibit the growth of the original controls and to encourage growth in the culture that had originally been retarded by the action of the light." They suggest that in many cases the mycelium may not be killed but rendered dormant for some considerable time.

STEVENS (21), in a group of fungi with which he says he obtained no interesting results from irradiations of 10, 30, and 50 seconds on corn meal agar "except stunting of colony and perhaps darkening and distortion or suppression of spore structures," lists: *Fusarium batatatis*, *F. conglutinans*, *F. niveum*, *F. oxysporum* (faint reddening on irradiation), *F. vasinfectum*, and *Fusarium* spp. (many). He also irradiated *Gibberella saubinetii*, which has as a conidial stage *F. graminearum*, and obtained with irradiations of 30 and 50 seconds distinct reddening of the mycelial tip, although he does not report the production of perithecia as in the case of *Colletotrichum lagenarium* (25).

JOHNSON (7) found that the aerial mycelium of *F. batatatis* Wr. collapsed, and that there was an increase in pigment within 24

hours after a 10-minute irradiation with Schumann rays. The collapsed mycelium "revealed a very granular protoplasm and the granules were relatively large." The mycelium was not entirely killed by a 25-minute exposure. Both macrospores and microspores occurred on controls but were more abundant after irradiation. Eight-day old cultures were less susceptible than those 5 days old. He found, also, that the ultraviolet transmitted by a Hanovia quartz mercury-arc lamp through cellophane (5790–2201 A.u.) produced no lethal effects on spores newly planted on agar, even if the dosage was as much as 50 minutes at 8 inches from the lamp. Three to 4-day old cultures given five daily exposures of 5–10 minutes through cellophane at 8–16 inches from the arc showed a "pronounced increase in pigment and a very noticeable suppression of growth." Zonation of the colony also occurred. The same types of reactions occurred to a less degree with filters transmitting to 2302 and 2536 A.u. With filters transmitting no lower than 2907 A.u. no variation was present after exposures as long as 3 hours at 8 inches.

RABINOVITZ-SERENI (13) worked with *Fusarium martii* and *F. poae*, and found that the spores of neither species were very resistant to the action of the ultraviolet emitted by a Gallois di Lione quartz mercury-vapor lamp at 20-cm. distance. Only a small percentage of those irradiated for 12 minutes or more germinated, and the germ tubes of these were abnormal.

Although much attention has been centered on the fungicidal action of short waves, some work has been published on the stimulating action of ultraviolet rays on fungi. NADSON and PHILIPPOV (9) reported a stimulating effect of ultraviolet rays on the growth of *Saccharomyces cerevisiae* and *Mucor genevensis* Lendn. In a subsequent paper (10) they reported work on four species of yeast and on *M. guilliermondii* Nad. & Phil. The fungi were exposed in petri dishes covered with a screen of thick glass or metal, in the center of which was a small round opening; the lamp used was a Bach quartz mercury-vapor model (100–120 volts, 3.8–4 amp.) transmitting from 2200 A.u. to 5700 A.u. Exposures of 10–20 minutes at 30 cm. from the arc were made 24 hours after inoculation of the plates with water suspensions of the organisms, and several days later it was found

that the part of the cultures submitted to direct radiation was dead, a sterile circle corresponding to the opening in the center of the screen. The rest of the colony, protected by the screen, continued to develop normally except in a narrow zone surrounding the sterile area, in which zone the yeast, influenced by the oblique rays, showed an intense multiplication and growth. Abnormal budding also occurred. In this "zone of stimulation" *M. guilliermondii* produced sporangia in great numbers although zygospores were lacking. In *M. genevensis*, on the other hand, it was the sexual reproduction which was induced in this area, the increased number of zygospores forming a black border to the sterile zone.

PORTER and BOCKSTAHLER (11) exposed *Colletotrichum lindemuthianum* and *Cephalothecium roseum* to wave lengths of 3630–3650 A.u. transmitted through a filter by a Cooper-Hewitt quartz mercury-arc lamp 60 cm. distant. They report "inhibition of growth and collapse and probably death of the aerial hyphae," and say: "In addition to these effects spore formation is inhibited sharply in *Cephalothecium* but on the other hand slightly accelerated in the case of *Colletotrichum*."

In the case of *Colletotrichum phomoides*, HUTCHINSON and ASHTON (6) were able to accelerate sporulation and retard growth by exposure of 3-day old cultures 12 cm. from the full mercury arc (Cooper-Hewitt) for 15 seconds to 2 minutes. Acervuli formed 24 hours after irradiation, with maximum development taking place in cultures irradiated for 30 seconds to 1 minute. Similar effects could be obtained by irradiating spores previous to planting on agar plates.

STEVENS, in works quoted and in subsequent papers (22, 23, 24), reports that he was able to induce pycnidia formation in such forms as *Camerosporium* sp. and in some strains of *Coniothyrium*, and to produce perithecia and acervuli in some strains of *Glomerella cingulata*. Doses approximately 300 times longer were required to kill conidia than were needed to induce perithecia of the last-named fungus. There was no such regularity in the induction of acervuli of *Glomerella cingulata* as in the case of perithecia. Both acervuli and perithecia failed to appear on irradiation of some strains.

DILLON WESTON (3) found that the retardation of growth result-

ing from exposure to ultraviolet wave lengths shorter than 400 mμ "may result subsequently in an acceleration of the development of the organism." Cultures of *Fusarium culmorum* were given one irradiation for different periods with light from a Hanovia sun-lamp. Although the primary reaction was one of retardation, subsequently the irradiated cultures sporulated abundantly whereas the non-irradiated cultures sporulated sparsely. In correlating these two facts he states, "it is assumed here that the initial retardation forces the organism to an earlier maturity."

The present paper attempts to make a comparative study of the effects of irradiation upon sporulation and upon certain physiological responses, such as color production and growth rate, in representative species from the described sections of the form genus *Fusarium*. All the sections are represented except section II, Macroconia, section IV, Submicrocera, and section V, Pseudomicrocera, of which no authentic cultures were obtained. All subsections are represented except Aquaeductuum under section Eupionnotes, which contains those species formerly placed in section Camptospora Wr. (**29**). The systematic arrangement followed is that given by WOLLENWEBER in his revised Delineata (**30**) and in the *Fusarium* monograph (**32**).

An attempt has been made to select, so far as possible, species which were typical for each section. Tests were made also with some of those having known perfect stages, in order to see whether irradiation would serve to stimulate production of the perithecial stage. More species were chosen from section Elegans (**28**, p. 28; **31**, p. 741) than from any other section, because this group contains more of the important plant parasites and because there is often greater difficulty in producing macrospores, and in distinguishing some of these forms, than is the case with species from the other sections.

Most of the cultures used were obtained from Dr. H. W. WOLLENWEBER or from Dr. O. A. REINKING at the time of the *Fusarium* conference in Madison, or are cultures brought to the conference by the writer and used in the studies (**34**) there. Credit is given in a footnote connected with table I for authentic cultures obtained from other sources.

Methods

Since, in the experiments with *F. cepae* and *Macrosporium* (14), irradiation through vitaglass covers transmitting to 2650 A.u. had proved most satisfactory, this glass was employed in most of the present experiments. Five-inch vitaglass squares were fitted as covers by molding paraffin rings about the edges of petri-dish bases. The lower half of the petri dish was held tightly against the vitaglass square until the paraffin had set, and then was slipped out. A little vaseline was mixed with the paraffin to make it less brittle when cold. These covers were sterilized with alcohol, and when dry were used to replace the ordinary petri-dish lid over the fungus culture previous to its exposure to ultraviolet.

In some of the experiments, a Stewart health-lamp equipped with therapeutic "A" carbons was used and operated on alternating current at 12 amperes through 110-volts resistance. This lamp is equipped with a Corex D filter which transmits traces between 2700 and 2800 A.u., feebly between 2800 and 2850 A.u., and absorbs none of the wave lengths above 2880 A.u. When the filter is not used the lamp gives some feeble transmission as low as 2300 A.u. Increased sporulation may be obtained with the Corex D filter, but in order to get the full effect of the lower wave lengths transmitted by the vitaglass it was necessary to remove the filter from the lamp. With the filter removed the heat produced tended to melt the paraffin rings if the plates were exposed for a long time at a distance of 40 cm. from the arc. For this reason most of the comparative studies were made with a Cooper-Hewitt quartz mercury-arc lamp, which does not produce so much heat. The mercury lamp was operated on alternating current at 4 amperes through resistance from a 110-volt line.

Plantings were made on potato dextrose agar, which had been found to be a very suitable medium for all species tested. For the purpose of these experiments it made little difference whether the inoculum used was a small piece of mycelium or a spore dilution, providing the same type of inoculum was used for control and test plates. The fungi were allowed to grow for four days before irradiation was begun. Some trials were made in which irradiation was begun on the second day, but the results were never so satisfactory.

If the cultures were much older than four days at the beginning of the tests, the mycelium often filled the plates before the end of the tests, thus hindering the measurement of growth rate.

Except in the experiments with various filters and in some of the direct exposures attempting to produce the sexual stage, the exposures were made through vitaglass covers 40 cm. from the unscreened mercury arc. Unless otherwise stated, all cultures were exposed for 15 minutes for each of three successive days. In all tests several plates were used for each exposure, and the average reading of many mounts made for total number of microspores and macrospores present, the results being recorded in terms of macrospore percentage of the total. Controls were held for an equal length of time in the diffused light of the laboratory, and the exposed plates were held in the same place as the controls when not actually under the lamp. All experiments were duplicated and questionable ones checked several times.

Results of irradiation through vitaglass

That not all species of *Fusarium* responded to irradiation with increased macrospore production, as did *Fusarium cepae*, is apparent from table I, which gives a summary of some of the experiments in which the cultures were irradiated through vitaglass for 15 minutes on each of three successive days. In most of the species exposed to ultraviolet there was an increase in total sporulation, if not in the percentage of macrospores produced. In some species, as *F. orthoceras* App. and Wr. var. *albidoviolaceum* (Dasz.) Wr. (Section XIV), the irradiated plates, although showing very few macrospores (fig. 1 *E*), actually had such an increase in microspore production as to produce an exposed pionnotes or at least marked zonation of the colony (fig. 1 *A*), while the control plates showed only aerial microspores and non-zonate mycelium. On the other hand, in forms like *F. stilboides* var. *minus* Wr. (Section XII), where microspores are usually lacking or scarce and where the percentage of macrospores present is therefore high in the control plates, the amount of macrospores in the irradiated plates was often increased to form sporodochia (fig 1 *B*) or a definite exposed pionnotes, as contrasted with few spores present in the controls. Usually increase in sporula-

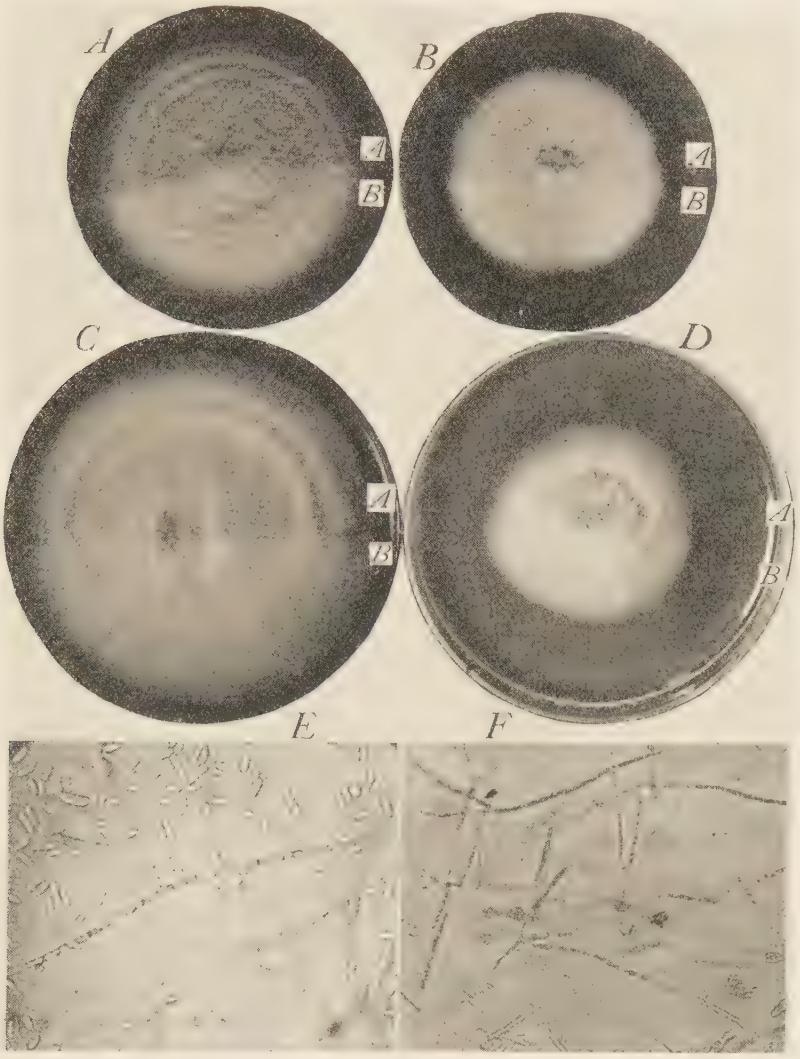


FIG. 1.—A–D, petri-dish cultures of *Fusarium* spp. showing effects of three daily 15-minute exposures to ultraviolet irradiation through vitaglass (2650–6690 A.u.), 40 cm. from quartz mercury arc: A, *F. orthoceras* var. *albidoviolaceum:* (a) irradiated portion showing zonate growth and deeper color due to sporulation; (b) control half (covered with photographer's black paper during irradiation); B, *F. stilboides* var. *minus:* (a) irradiated portion showing sporodochia; (b) control half; C, *F. nivale:* (a) irradiated portion showing more powdery appearance and zonation; (b) control half; D, *F. dimerum:* (a) irradiated portion showing exposed pionnotes; (b) control portion; E, *F. orthoceras* var. *albidoviolaceum* showing scarcity of macrospores after three daily 15-minute exposures through vitaglass; F, *F. argillaceum* showing injured spores and mycelium and chlamydospores produced on old portion of colony which had received three daily 15-minute exposures through vitaglass.

TABLE I

SUMMARY OF EXPERIMENTS SHOWING EFFECTS OF THREE SUCCESSIVE 15-MINUTE EXPOSURES TO ULTRAVIOLET TRANSMITTED THROUGH VITAGLASS (LOWER LIMIT OF TRANSMISSION, 2650 A.U.)

SECTION	SPECIES OF FUSARIUM	PERCENTAGE MACROSPORES		OTHER DIFFERENCES
		IRRADIATED	CONTROL	
1. Eupionnotes	F. dimerum Penz. (*R*)*	0.0	0.0	More microspores and deeper color on exposed plates
	F. merismoides Cda. var. majus Wr. (Syn. F. betae (Desm.) Sacc. (*W*)	10 9	7 7	Total sporulation (macro- and microspores) greater on tests
2. Macroconia	No representative			
3. Spicarioides	F. decemcellulare Brick. (*R*)	1.0	−1.0	More microspores on exposed
4. Submicrocera	No representative			
5. Pseudomicrocera	No representative			
6. Arachnites	F. nivale (Fr.) Ces. (*D–L*)	68.5	53.5	Exposed culture more powdery
7. Sporotrichiella	F. sporotrichioides Sherb. (*B*)	60.7	32.8	Thicker pionnotes on exposed plates
8. Roseum	F. anguioides Sherb. (*R*)	6.0	−1.0	
9. Arthrosporiella	F. semitectum B. & Rav. (*W*)	23.7	19.4	
	F. semitectum var. majus Wr. (*R*) (Syn. F. incarnatum (Rob) Sacc.)	94.3	64.1	Thicker pionnotes on exposed plates

* Letters in italics refer to source of the culture as follows:

B, Writer's culture
C, G. H. COONS
D–L, A. S. DAHL through G. K. K. LINK
G, W. W. GILBERT
H, L. L. HARTER
J, HELEN JOHANN
R, O. A. REINKING
S, M. SHAPOVALOV
T–N, J. J. TAUBENHAUS through A. G. NEWHALL
W, H. W. WOLLENWEBER
W–J, WOLLENWEBER through HELEN JOHANN
Wis, Wisconsin laboratory
W–L, Wisconsin laboratory through G. K. K. LINK

TABLE I—*Continued*

10. Gibbosum	F. equiseti (Cda) Sacc. f. 1 Wr. (*R*) (Syn. F. ossicolum (B & C) Sacc.)	37.7	27.0	Exposed pionnotes on irradiated plates
	F. scirpi L. & F. var. acuminatum (E. & E.) Wr. (*R*)	89.0	50.0	Very few spores in control; more plentiful on exposed
	F. scirpi L. & F. var. longipes (Wr. & Rg.) Wr. (*R*)	51.7	38.1	
	?F. scirpi L. & F. var. compactum Wr. (*B*)	75.0	0.0	See text
	?F. scirpi L. & F. var. compactum Wr. 2nd test after spores were once obtained by irradiation	71.4	69.0	See text
11. Discolor ss. Neesiola	F. heterosporum Nees. var. paspalicola (P. Henn.) Wr. (*W*)	1.8	0 to −0.5	Total sporulation greater after irradiation
ss. Trichothecioides	F. trichothecioides Wr. (*B*)	5.1	−1.0	A good many 2-septate spores on irradiated plates
ss. Saubinetii	F. macroceras Wr. & Rg. (*R*)	−0.5	−0.5	Total sporulation greater on exposed plates
	F. culmorum (W. G. Sm.) Sacc. (*W–J*)	83.2	0.0	No microspores on controls
	F. sambucinum Fuckel (*S*)	93.8	91.0	Irradiated plates and controls look alike
	F. sambucinum Fuckel (*B*)	95.0	0.0	No microspores on controls; few spores on treated plates
12. Lateritium	F. stilboides var. minus Wr. (*R*) (Syn. F. fructigenum Fr. var. maius Wr. f. 1 Wr. & Rg.)	93.2	81.8	Spores rare in control; plentiful in definite pionnotes on exposed plates

TABLE I—*Continued*

Section	Species of Fusarium	Percentage macrospores		Other differences
		Irradiated	Control	
13. Liseola	F. neoceras Wr. & Rg. (*R*)	35.2	3.7	Greater sporulation on exposed plates
	F. moniliforme Sheld. (*R*)	−1.0	0	" " " " "
	F. " " var. subglutinans Wr. & Rg. (*R*)	6.0	3.2	" " " " "
	F. moniliforme Sheld. var. erumpens Wr. & Rg. (*R*)	−1	−0.5	" " " " "
14. Elegans ss. Orthocera	F. orthoceras App. & Wr. var. longius (Sherb) Wr. (*W*)	4.4	3	
	F. orthoceras var. albidoviolaceum (Dasz) Wr. (*C*)	−1	−0.5	Exposed pionnotes and many microspores on irradiated plates; no pionnotes and few microspores on controls
	F. orthoceras var. albidoviolaceum (Syn. F. asclerotium) (*strain from W*)	1.7	0	Total sporulation greater on irradiated plates
	F. orthoceras var. triseptatum Wr. (*R*)	−1	−1	Thin pionnotes on exposed plates; no pionnotes and few microspores on controls
	F. bostrycoides Wr. & Rg. (*R*)	−0.5	−0.5	Exposed culture more powdery
	F. conglutinans Wr. (*Wis*)	−1.0	0 0	Total sporulation greater on irradiated plates
	F. " " var. callistephi Beach (*C*)	7.4	−0.5	Exposed pionnotes on irradiated plates; not on controls
ss. Constrictum	F. bulbigenum Cke. et Mass. f. 1 Wr. (Syn. F. lycopersici) (*W*)	−1	0	Thicker pionnotes and deeper color than controls
	F. bulbigenum Cke. et Mass. f. 1 Wr. (Syn. F. lycopersici) (*strain 2-W*)	4.3	3.7	As above; strains from 2 other sources acted the same
	F. bulbigenum var. batatas Wr. (Syn. F. batatatis Wr.) (*H*)	4.8	0.8	Relatively few spores in controls; many in treated plates

TABLE I—*Continued*

ss. Oxysporum	F. oxysporum Schlect. (*R*)	−1	−0.5	
	F. " " (*2nd strain S*)	2 3	−0.5	Total sporulation greater on irradiated plates
	F. " " f. 1 Wr. (*W*) (Syn. F. euoxysporum Wr.)	2.2	0	As above
	F. oxysporum f. 2 Wr. (*S*) (Syn. F. hyperoxysporum Wr.)	2	−1	Thinner pionnotes on controls; 2 strains from Harter acted same way
	F. oxysporum f. 3 Wr. (Syn. F. cubense E.F.S.) (*2 strains R*)	Strain A, 8 Strain B, 1	0 0	No pionnotes on control; thin one on irradiated plates
	F. oxysporum f. 5 Wr. (*R*) (Syn. F. oxysporum var. nicotianae Johns)	69	28.3	Pionnotes thinner and covered with mycelium in controls
	F. oxysporum var. aurantiacum (Lk.) Wr. (Syn. F. aurantiacum) (*W*)	7.6	0	Total sporulation greater on exposed plates
	F. cepae Hanz. emend. Link & Bail.	51.0	5.0	Exposed pionnotes on irradiated plates
	F. vasinfectum Atk. (*G*)	1	1	Total sporulation greater
	F. " " f. 1 Wr. (Syn. F. vas. var. inodoratum) (*W*)	96.9	83	More mycelium in controls
	F. vasinfectum var. lutulatum (Sherb.) Wr. (*R*)	55.5	42.4	No aerial mycelium on exposed plates
	F. vasinfectum var. zonatum (Sh) Wr. (Syn. F. zonatum) (*W*)	28	4.9	Pionnotes thicker on exposed plates
	F. vasinfectum var. zonatum f. 1 (Lk. & Bail.) Wr. (*B*)	81.9	69	" " " " "
	F. vasinfectum var. zonatum f. 2 (Lk. & Bail.) Wr. (*B*)	59.0	51.2	" " " " "
	F. redolens Wr. (*W*)	83.9	53.4	" " " " "
15. Martiella	F. coeruleum (Lib) Sacc. (*S*)	96.8	0	No spores of any kind on controls; pionnotes on irradiated plates
	F. solani App. & Wr. var. medium Wr. (Syn. F. malli Taub.) (*T–N*)	59.1	3.4	Exposed pionnotes on irradiated plates
	F. javanicum Koord. (*R*)	39.9	17.0	See text
16. Ventricosum	F. argillaceum (Fr.) Sacc. (*W–J*)	73.2	65.9	Exposed plates show appressed moist mycelium, much stunting and many chlamydospores

tion was noticeable macroscopically in production of sporodochia or pionnotes, or at least in greater intensities of color in mycelium, stroma, and spore mass. In *F. nivale* (Fr.) Ces. (Section VI), although the increase in macrospore percentage was not marked, the treated colonies were much more powdery owing to the increased sporulation (fig. 1 *C*). In such species as *F. dimerum* Penz. (Section I), where macrospores do not occur, an increase in the microspore production resulted in an exposed pionnotes on irradiated plates (fig. 1 *D*), but there was no evident increase in septate over continuous spores. This increase was accompanied by an intensification of the mycelium color. RABINOVITZ-SERENI (12) reports such an intensification of colors in cultures of *F. poae* exposed to ordinary blue light, but does not report any associated increase in quantity of spores present.

In some species the macrospore percentage in exposed plates was more than double that in the control plates. In *F. neoceras* Wr. and Rg. (Section XIII) there was an increase from 3.7 to 35.2 per cent; *F. cepae* (Section XIV) increased from 5 to 51 per cent macrospores; and *F. solani* App. and Wr. var. *medium* Wr. (Syn. F. *malli* Taub.) (Section XV), which had given very few normal macrospores in ordinary laboratory culture, showed an average of 59.1 per cent macrospores after irradiating for 15 minutes through vitaglass on three successive days. In most species the response to irradiation was relatively consistent although the percentages varied. For instance, it will be noted (table I) that the average percentage of macrospores present in the *F. sporotrichioides* Sherb. (Section VII) plates after irradiation was 60.7, not quite double that in the control plates, whereas in the experiments summarized in table III the average macrospore production under vitaglass was 51.7 per cent, or five times that of the controls; but in all experiments there was a decided increase in percentage of macrospores. The average percentage for all the experiments with *F. vasinfectum* Atk. var. *zonatum* (Sh.) Wr. (Section XIV), when treated to three successive 15-minute exposures under vitaglass, was 12.1 as against an average of 3 per cent in the controls. Individual tests varied from 8 per cent in the irradiated plates with 0.5 per cent in the controls to 46.7 per cent in the irradiated plates with 5 per cent in the controls. Out of

ten such experiments with this species, each involving several plates, the percentage on the irradiated plates was usually five or six times that from the controls, was never less than three times, and was only once as high as ten times that obtained in the controls.

On the other hand, in an equal number of experiments with *F. conglutinans* Wr. and with *F. bulbigenum* Cke. et Mass. forma I Wr. (syn. *F. lycopersici* [Sacc.] Wr.) and others, the percentage was never increased by as much as 1 per cent under this treatment. The tendency to show definite, unmistakable macrospore production when exposed to the ultraviolet rays transmitted by a quartz mercury arc through vitaglass seemed to be characteristically present or absent for any one of the species tried in these experiments with but few exceptions. The exceptions were *F. scirpi* L. and F. var. *acuminatum* (E. and E.) Wr. (Section X), *F. javanicum* Koord. (Section XV), and *F. argillaceum* (Fr.) Sacc. (Section XVI).

In *F. scirpi* var. *acuminatum* most of the experiments showed striking increase in macrospore content after irradiation, but in two experiments the increase was slight, and in one the controls averaged a higher percentage than the treated plates. In this latter case, however, the number of spores actually present on the control plates was so small as to require examination of a great many mounts before sufficient spores were obtained for counting, whereas in the exposed plates the spores were plentiful although low in macrospore count. Where spores are rare, one would expect a high macrospore count with *F. scirpi* var. *acuminatum*, for even when not in "hochkultur" it produces few microspores. However, the macrospores in control cultures made from subcultures of a strain which had been carried long in artificial culture were often small 3–4 septate spores as contrasted with the well shaped normal 5-septate spores of the irradiated cultures.

F. javanicum is another species which ordinarily produces very few microspores, but the strain which had been received from REINKING in 1924 had been carried in artificial culture since that time and was producing a high percentage of 1-septate and small spores. In some tests the irradiated plates, in contrast to the controls (fig. 2 *A*), produced a fair percentage of normal 4–5-septate spores (fig. 2 *B*) but in other experiments irradiation had little effect. Apparent-

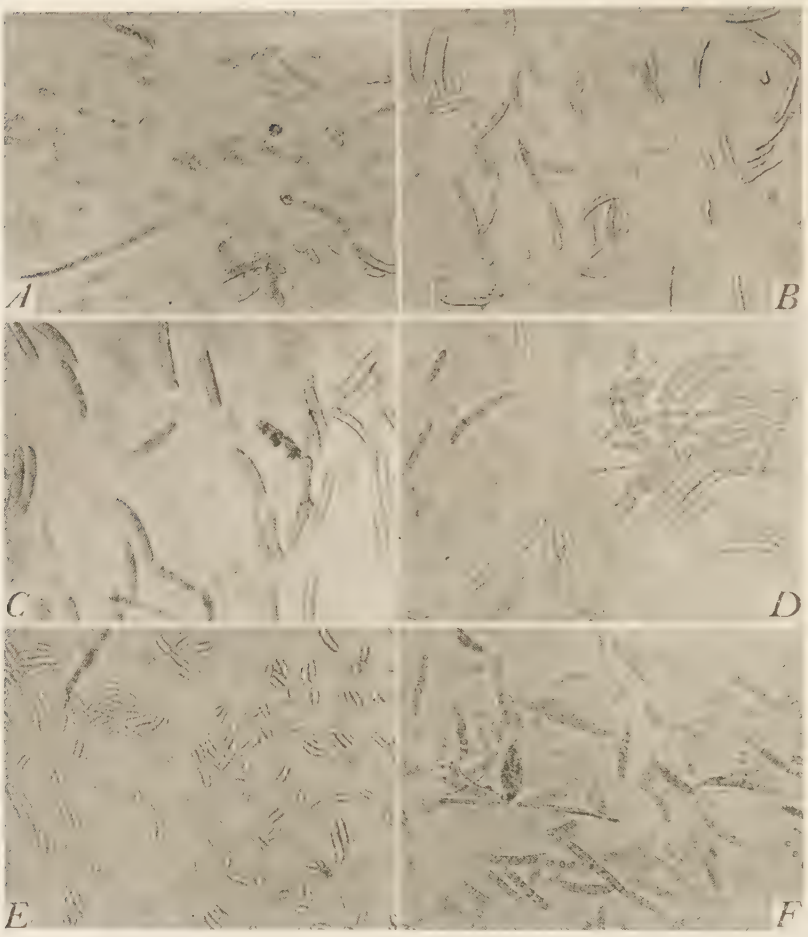


FIG. 2.—Effects of ultraviolet irradiation on cultures of *Fusarium* spp., placed 40 cm. from quartz mercury arc: A, *F. javanicum* control showing predominance of microspores; B, *F. javanicum* irradiated through vitaglass for three daily 15-minute periods with resultant increase in macrospore percentage; C, macrospores of *F. coeruleum* produced by three daily 15-minute exposures through vitaglass. D–E, *F. vasinfectum* var. *zonatum:* D, abundance of macrospores produced as result of three daily 15-minute irradiations through Corning Corex A filter 980 A (2300–6690 A.u.); E, control showing scarcity of macrospores; F, plasmolysis of protoplasm in spores as result of three daily 15-minute exposures direct to the lamp (2230–6690 A.u.).

ly macrospore production by this method cannot be predicted for *F. javanicum.*

In the case of *F. argillaceum,* the macrospore percentage on the irradiated plates was usually a little higher than on the control plates, but sometimes this did not hold true. It was found that if the mounts were made only from the new growth that occurred during the 24 hours following irradiation, the macrospore percentage was always higher and the spores normal; but if the mounts were made only from the portion of the colony that had actually been irradiated, there was considerable variation in results. The spores (fig. 1 *F*) present in this irradiated area were usually abnormal in shape, contained refractive particles apparently in part fat and glycogen, or were plasmolyzed or breaking down, so that even though present they could not be used for study of normal spores. In all experiments great care was taken to secure uniform spore mounts from all over the colony, but in the case of *F. argillaceum* this was always more difficult, owing to the toughness and irregular growth of the stunted colony after irradiation. Even those rays transmitted by vitaglass usually resulted in disruption of spores and mycelium of *F. argillaceum,* and in retarded, abnormal growth. Lengthening the period of exposure to one half-hour resulted generally in such colony stunting (measured by radial increase and amount of aerial mycelium) and death of spores that recovery did not take place in the 24 hours succeeding exposure, with the result that the macrospore percentage was less than that of the controls at the close of the experiments. Shortening exposure to one 15-minute period gave better production of macrospores. The irradiation usually resulted in the production of many chlamydospores, whereas the control plates had very few; with longer exposures the growth often was largely chlamydospores.

The most outstanding usefulness of irradiation with ultraviolet is in inducing the production of spores by strains which have not been sporulating; and the method is an advantageous one, for if the species is one responsive to ultraviolet treatment macrospores are readily obtained in a very short time. Even the production of microspores is extremely helpful in such studies, for purification of the

culture by obtaining single spore strains and for help with taxonomic identification. One strain (tentatively placed in *F. scirpi* L. and F. var. *compactum* Wr. of Section Gibbosum), isolated from decayed onion bulbs, had never produced spores in artificial culture until irradiated, when normal spores suitable for taxonomic study appeared on the experimental plates. While the treated plates had many spores (75 per cent macrospores) the control plates were still without spores of any kind. In subsequent experiments with this fungus, when the inoculum was taken from the pionnotal stage which now occurred in subcultures made from the previously treated plates, irradiation failed to bring the percentage of macrospores noticeably higher than the controls which were already producing abundant macrospores, as shown in table I.

A culture of *F. coeruleum* (Lib.) Sacc., which likewise had failed to produce spores of any kind in artificial culture, was induced to produce abundant macrospores (fig. 2 *C*) with irradiation, while the controls remained sterile. A strain of *F. sambucinum* Fuckel, which was in "abkultur," was made to produce macrospores by irradiation although the controls produced no spores; and similarly, irradiated plates of *F. culmorum* (W. G. Sm.) Sacc. produced 83.2 per cent macrospores when no conidia could be found in the control plates. In the case of the latter species it was a matter of hastened spore production, for a week later both control and treated plates showed spores. Yet even after this interval of time effects of irradiation were evident, for the control plates averaged from 40 to 56 per cent macrospores and the exposed plates from 65 to 70 per cent. DILLON WESTON (3) also found hastened sporulation in irradiated cultures of this species, but the increase in macrospore percentage here given would seem to indicate that the effect of irradiation is something more than earlier maturation. Preliminary observations of irradiated *Fusarium* spp. indicate that changes in the composition of the mycelium may be associated with the assumption of sporulation following irradiation.

It is interesting to note in table I, and in subsequent tables, that the species which responded to treatment with marked increase in macrospore production are the saprophytes and decay-producing organisms and not the vascular parasites. The only vascular para-

site which showed appreciable increase was *F. oxysporum* Schlect. forma 5 Wr. (syn. *F. oxysporum* Schlect. var. *nicotianae* Johns.), whose control plates averaged 28.3 per cent macrospores while the irradiated plates averaged 69 per cent. This culture of *F. oxysporum* f. 5 is a subculture of Dr. REINKING'S culture no. R119 of *F. oxysporum* var. *nicotianae*, which is the strain represented by WOLLENWEBER in Delineata 1012 (1930 Supplement) (**30**) and described by REINKING and WOLLENWEBER in Tropical Fusaria. In the latter reference (**15**, p. 192) the authors state in text and footnote:

> *Fusarium oxysporum* var. *nicotianae* is the cause of a tobacco wilt in the United States, but the pathogenicity of the soil fungus described here has not yet been proved. . . . . Under such circumstances the fungus described here, as isolated from Honduras soil and identified as *F. oxysporum* var. *nicotianae* may touch the border line or may be even a widespread saprophytic soil form of the banana wilt *Fusarium, F. cubense.*

Incidentally the strains here tested of the banana parasite, *F. cubense* E. F. S., also obtained from REINKING, gave little response to ultraviolet. Whether or not this variation in response of two large groups of *Fusarium* species will prove constant in tests with other species of these groups, and whether such differences will prove significant in separating vascular parasites from non-vascular strains, awaits further experimentation. It is possible that more response to light might be expected from organisms which were more often exposed to light in nature on the surface of decaying material, etc., than from the vascular parasites which are spread through the soil, less exposed to sunlight, and also less dependent upon spores for their spread. At least, in contrast with the response of the vascular parasite group, irradiation was most positive in its effect on the production of macrospores in the following sections: Sporotrichiella, Gibbosum, Discolor subsection Saubinetii, Martiella, and Elegans subsection Oxysporum (except vascular parasites).

It was found, as in the previous work with *F. cepae* (**14**), that irradiation of responsive species on several days in succession was better than one longer exposure. One day's exposure with responsive species gave results, but not so many macrospores developed as were obtained by several irradiations. Fifteen minutes under vitaglass on each of three successive days was a good average period for

this purpose; under some of the filters transmitting shorter wave lengths a shorter period of time was favorable. Increasing the exposure beyond 15 minutes sometimes resulted in a still greater increase in sporulation, sometimes not. Plates of *F. decemcellulare* Brick., given three 30-minute exposures, produced 3.1 per cent macrospores as against 1 per cent after three successive 15-minute exposures and in the controls; with half-hour exposures of *F. heterosporum* Nees. var. *paspalicola* P. Henn. it was possible to get 4.5 per cent as against 1.8 per cent under 15-minute treatments. But these differences are slight and it was usually found that if treatment for 15 minutes under vitaglass was not effective, longer exposures were seldom warranted, and in some fungi, such as *F. argillaceum*, were actually lethal or detrimental to normal vegetative and reproductive development.

Multiplying the numbers of days of exposure was often more effective for conidial production than lengthening the period of a single exposure; but in species entirely unresponsive to three 15-minute treatments, even this increase in number of irradiations failed to produce spores. Table II shows the results of three and of six daily 15-minute exposures with several species of *Fusarium*. In all of the experiments shown in this table several plantings were made to the plate, and the readings made on the seventh day were from different colonies on the same plates as those read for the fourth-day results. The readings on the fourth day were, of course, made before irradiation on that day. In a few species, such as *F. vasinfectum* var. *zonatum* forma 2 and *F. oxysporum* Schlect. forma I Wr. (syn. *F. oxysporum* var. *euoxysporum* Wr.), there was noticeable increase in the six exposures, but in some of them (*F. javanicum* and *F. argillaceum*, for instance) the spore count was less at the end of the six days.

An attempt was made to see whether a cumulative effect could be obtained in some of the species which had failed to respond with macrospore production. For the purposes of this study, spores from plates which had already been irradiated were used as inoculum for new petri-dish cultures to be irradiated. The tests with *F. orthoceras* App. & Wr. will serve to illustrate the results. A strain which had been producing few spores in the stock tubes was used in the first experiments, and the usual tests involving three 15-minute

exposures resulted in a deepening of the color of the colony and the production of more microspores than the controls. Less than 1 per cent macrospores were present in controls and test plates alike. A duplicate experiment was run with three half-hour exposures which

TABLE II

EFFECTS ON SPORULATION OF FUSARIUM SPECIES OF THREE AND OF SIX DAILY 15-MINUTE EXPOSURES TO ULTRAVIOLET RAYS TRANSMITTED THROUGH VITAGLASS

SPECIES OF FUSARIUM	EXPOSED (PERCENTAGE MACROSPORES)		CONTROL (PERCENTAGE MACROSPORES)	
	3 DAYS	6 DAYS	3 DAYS	6 DAYS
F. argillaceum (Fr.) Sacc. (*W-J*)*	67.3	42.7	62.6	.....
F. bulbigenum Cke. & Mass. f. 1 Wr. (*C*)	5.4	7.0	6.7	6.0
F. conglutinans Wr. (*Wis*)	0 to −0.5	3.9	0.0	2.0
F. " " var. callistephi Beach (*C*)	7.4	3.8	1.0	1.0
F. decemcellulare Brick. (*R*)	2.2	3.1	0.6	1.0
F. heterosporum Nees. var. paspalicola (P. Henn.) Wr. (*W*)	2.0	4.5	−0.5	−0.5
F. javanicum Koord. (*R*)	39.9	9.2	17.0	17.0
F. macroceras Wr. & Rg. (*R*)	−0.5	−0.5	−0.5	−0.5
F. moniliforme Sh. var. maius Wr. & Rg. (*R*)	0.6	1.6	1.0	1.0
F. orthoceras App. & Wr. (*W*)	1.0	1.0	−0.5	−0.5
F. " var. albidoviolaceum (Dasz) Wr. (*W*)	1.7	0.9	0.0	0.0
F. oxysporum Schlect. (*R*)	−1.0	−1.0	−0.5	−0.5
F. " forma 1 Wr. (*W*)	2.2	12.7	0.0	0.0
F. " " 2 Wr. (*S*)	3.0	1.9	−1.0	1.0
F. " " 3 Wr. (*R*)	−1.0	−1.0	0.0	0.0
F. " var. aurantiacum (Lk) Wr. (*W*)	7.6	7.0	0.0	−0.5
F. redolens Wr. (*W*)	83.9	86.3	53.4	55.0
F. scirpi L. & F. var. acuminatum (E. & E.) Wr. (*R*)	64.7	99.3	78.8	79.6
F. semitectum B. & Rav. (*W*)	28.9	22.1	18.5	16.6
F. vasinfectum Atk. (*B*)	2.0	4.8	3.8	3.5
F. " strain 2 (*G*)	1.0	1.6	1.0	1.0
F. " forma 1 Wr. (*W*)	96.9	98.2	83.0	81.0
F. " v. zonatum (Sh) Wr. f. 2 Lk. & Bail. (*B*)	56.8	78.6	55.0	55.0

* Letters in italics refer to source of the culture as shown in footnote, table I.

gave results detectable from the 15-minute plates only in a slightly deeper color. Mounts of spores from the irradiated plates of these two sets of tests, and also from their controls, were then used as inoculum for petri-dish cultures, which after sufficient growth were given three daily 15-minute treatments with ultraviolet transmitted through vitaglass. Some of the previously irradiated plates and

their controls, one each of which had been used for the inoculum, were retained for comparison at the conclusion of the new tests. A control for the whole test was made by inoculating plates with mounts from the subcultures of the original unirradiated culture.

At the conclusion of the three days, the newly irradiated plates (*A*), obtained from the inoculum previously given three 30-minute exposures, showed a thick zonate light ochraceous salmon (**16**) pionnotes with many more spores than the controls, and with a deeper pionnotes than the plates from which the inoculum was taken; but there were still less than 1 per cent macrospores. The newly irradiated plates (*B*), whose inoculum came from the cultures previously given a 15-minute exposure period, also showed more spores than the control and than their inoculum plates; but there was more aerial flesh pink mycelium and a little less exposed pionnotes than on the *A* plates. Here again the microspores were more than 99 per cent of the total spore content. The newly exposed plates (*C*), with inoculum taken from the former control cultures, looked like the *B* plates and had a similar spore content. All unirradiated controls showed appressed moist or aerial white mycelium with no visible pionnotes and with a spore composition made up almost entirely of microspores.

A repetition of the experiment using plates *A*, *B*, and *C* for inoculum served to produce a slightly thicker pionnotes in all irradiated plates but no macrospore increase. The second subculture of the original unirradiated culture showed a very slight increase in spore production but no real pionnotes. Evidently the increase in spore content after irradiation was a little more than that ordinarily obtained by making subcultures from spores instead of from mycelium, for in every case treatment increased the total spore production more than did simple subculture by spore inoculum from the control. However, the tests failed entirely to produce a cumulative effect in percentage of macrospores present.

Similar results were obtained with strains of *F. conglutinans* and with *F. bulbigenum* forma 1 (*F. lycopersici*) which had also been unresponsive to early irradiation tests. In other strains of *F. bulbigenum* forma 1, which had been producing a few macrospores in stock tubes, repeated irradiation resulted in an increase in the total

spore production and in the number of macrospores present, but no appreciable increase in percentage. Thus the response seemed consistent regardless of whether the fungus treated was in "abkultur" or "hochkultur."

Results of irradiation through various filters

A distance of 40 cm. from the arc was found most advantageous for spore production. If this distance was shortened much, the factor of heat from the lamp entered in, and if it was lengthened much, the loss of the shorter rays became a factor. Although the distance from the arc and the length of exposure most favorable seemed to be about the same for most species tried, it was found that maximum production of spores was not induced in every strain when a vitaglass filter was used. This is brought out in figures 5 and 6 and in table III, which gives wave lengths transmitted by each filter and by the lamp itself. Figure 3 gives the spectrograms of the quartz mercury arc direct and through the various filters.

In the experiments with the filters, all plates were exposed for 15 minutes on each of three successive days at 40 cm. from the arc, but with the filter inserted in the lamp between the arc and the culture. During exposure the petri-dish cover was removed. Direct exposure was made without any filter in the lamp. Lids were removed from the controls also, for 15 minutes each day during exposure of the other plates. When not under the lamp, all plates were kept covered in diffuse light in the laboratory.

Although for most species tried, the vitaglass, transmitting wave lengths as short as 2650 A.u., gave maximum or very good results in spore production, maximum results with *F. vasinfectum* var. *zonatum* were always obtained by using Corex filter 980 A. (fig. 2 *D*), whose lower limit of transmission is 2300 A.u. Use of this filter gave as much as two or three times as high a percentage of normal macrospores as did the use of the vitaglass filter, although the latter filter induced three to eight times as high a percentage of macrospores as was present in the unirradiated plates (fig. 2 *E*). Use of Corex filter 986 A gave slightly better results with *F. vasinfectum* var. *zonatum* than did the vitaglass, and, although some normal spores as well as injured spores (fig. 2 *F*) occurred when the cultures were irradiated

TABLE III

EFFECT ON MACROSPORE PRODUCTION OF EXPOSING (AT 40 CM. FROM ARC FOR 15 MINUTES ON EACH OF THREE SUCCESSIVE DAYS) FUSARIUM SPECIES TO ULTRAVIOLET RAYS EMITTED BY QUARTZ MERCURY ARC THROUGH VARIOUS FILTERS

SPECIES OF FUSARIUM	CONTROL (DIFFUSE DAYLIGHT)	NOVIOL C 4880–6690 A.U.	G 586 AW 3334–3690 A.U.	FILTER NO. 1 3020–6690 A.U.	VITAGLASS 2650–6690 A.U.	COREX 986 A 2535–4340 A.U.	COREX 980 A 2300–6690 A.U.	DIRECT. 2230–6690 A.U.
F. decemcellulare Brick. (*R*)† (section Spicarioides)	Macro. rare	Macro. rare	Macro. rare	Macro. less than ½% *	Macro. ½% *	Macro. less than ½% *	Less than ½% *	No macro.; few micro. (4-under agar surface)
	(8 mm.)‡	(9)	(8)	(8)	(8)	(6.5)	(5.5)	
F. nivale (Fr.) Ces. (*D–L*) (s. Arachnites)	59%§	61.3%	63.5%	61.8%	75.2% *	73.1% *	69% *	76.7%
	(10)	(10)	(10)	(10)	(10)	(10)	(10)	(5)
F. sporotrichioides Sh. (*B*) (s. Sporotrichiella)	10.9%	10.5%	9.6%	11.8%	51.7% *	53.5% *	34.3% *	16%
	(15.5)	(17)	(16.5)	(16)	(16)	(13.5)	(12.5)	(7.5)
F. semitectum B. & Rav. (*R*) (s. Arthrosporiella)	18.4%	18.9%	18.1%	16.8% *	18.8% *	20% *	25.3% *	39.5% (?) (3-on surface 12-beneath)
	(15)	(16)	(15)	(15)	(15)	(12)	(13)	
F. scirpi L. & F. var. acuminatum (E. & E.) Wr. (*R*) (s. Gibbosum)	51.9%	55%	64.6%	62.2%	85.7% *	71.1% *	71.9% *	Macro. & micro. rare (2)
	(17.5)	(22.5)	(17)	(18.5)	(17)	(12)	(12)	
F. sambucinum Fuckel (*B*) (s. Discolor)	No spores	No spores	No spores	Spores rare, mostly macro. *	95% *	No spores	No spores	No spores
	(8)	(7)	(8)	(7)	(7)	(4)	(5)	(1)

* Total sporulation (macrospores and microspores) greater than controls.
† Letters in italics refer to source of culture as given in footnote, table I.
‡ Figures in parentheses express radial increase in mm. during 3 days following first exposure.
§ All percentages express percentage of macrospores present

TABLE III—*Continued*

F. neoceras Wr. & Rg. (*R*) (s. Liseola)	1.9% (14.5)	2.5% (14)	2.5% (15)	6.3% * (14.5)	22.7% * (14)	8.6% * (14)	4.4% * (14)	Macro. & micro. rare (11)
F. orthoceras App. & Wr. (*W*) (s. Elegans s.s. Orthocera)	No macro.; very few microspores (14)	Like control (14)	Like control (14)	No macro.; thin pion-notes * (15)	Less than 1%; * thin pion-notes (14)	Rare; * thin pion-notes (12)	Like 986 A * (8.5)	All spores rare (6.5)
F. conglutinans Wr. (*Wis*) (Elegans-Orthocera)	No macro. (10)	No macro. (10)	Macro. rare (10)	Macro. rare * (10)	Less than 1% * (10)	Rare * (9)	No macro. * (9)	No macro.; very few micro. (5)
F. bulbigenum Cke. & Mass. (*W*) (Elegans-Constrictum)	3 3% (14 3)	1.7% (15)	3.3% (15)	2.7% (15)	8.1% * (13)	17.2% * (12)	11.4% * (11)	12.8% (6-mostly beneath agar)
F. vasinfectum Atk. var. zonatum (Sh) Wr. (*W*) (Elegans-Oxysporum)	5.6% (13.3)	5.1% (13.5)	5.6% (13.3)	10 9% (14)	17.5% * (14 4)	19.2% * (12)	37.7% * (13)	17.6% (8.2-mostly under agar)
F. javanicum Koord. (*R*) (s. Martiella)	3.4% (16)	4.2% (15.5)	5.6% (17)	6% * (15.5)	10.2% * (14)	9.2% * (10)	8.6% * (8)	Spores rare; many shattered (2 above agar 5 mm. total)
F. javanicum Koord. var. theobromae (App. & Strk.) Wr. (*R*) (s. Martiella)	Macro. rare (17)	Macro. rare (16)	Macro. rare (16)	Macro. rare (17)	Less than 1% * (12)	Less than 1% * (8)	More than vitag. but not 1% * (9)	Like 980 A but spores abnormal (3)
(F. coeruleum (Lib) Sacc. (*S*) (s. Martiella)	No spores (6)	No spores (5.5)	No spores (6)	Few spores * 95.6% (5)	Many spores * 99.4% (5)	Fewer spores than vitag. * 91.9% (3)	Like 986 A * 85.6% (3.5)	No spores; mycelium empty or granular (1)

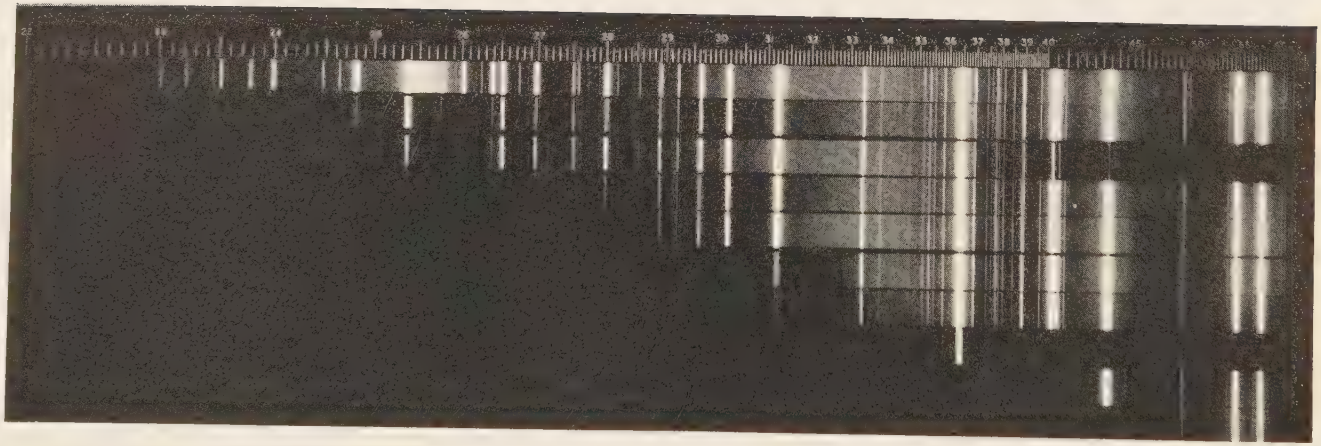


FIG. 3.—Spectrograms of quartz mercury arc direct and through various filters: A, direct arc (2230–6690 A.u.); B, Corning Corex A (980 A), (2300–6690 A.u.); C, Corex red-purple (986 A), 2535–4340 A.u.; D, vitaglass (2650–6690 A.u.); E, petri dish (2800–6690 A.u.); F, glass filter no. 1 (3020–6690 A.u.); G, glass filter no. 6 (3120–6690 A.u.); H, Corning (G 586 AW), (3334–3690 A.u.); I, Noviol A (4340–6690 A.u.); J, Noviol C (4880–6690 A.u.). Some faint lines showing lowest limits of transmission are lost in reproduction.

directly, usually the percentage was less than with any of the three filters transmitting as low as 2650 A.u.

The peak of macrospore production for *F. sporotrichioides* occurred consistently with the use of Corex filter 986 A (fig. 4 *A*), with which the shortest wave length transmitted was 2535 A.u. Although there was little difference between the results with this filter and those with vitaglass, the small advantage in number of macrospores produced was always on the side of the Corex 986 A (table III). Fewer sporotrichial, pyriform conidia (**19,** p. 183) occurred with 986 A than with vitaglass. When the fungus was exposed to the still shorter wave lengths transmitted by Corex filter 980 A, there was a decided drop in macrospore production but also in the sporotrichial conidia, the microspores present being ovate rather than pyriform. With filters transmitting no shorter waves than 3020 A.u. there was no noticeable change from the control plates, all of which produced few macrospores and a high percentage of pyriform conidia (fig. 4 *B*). *F. bulbigenum* Cke. and Mass. ran a much more evident peak under Corex filter 986 A than did *F. sporotrichioides*, for in every experiment the spore production here was much better and the macrospores produced (fig. 4 *C*) were of better shape than under vitaglass (fig. 4 *D*) and Corex 980 A. Sporulation on the control plates was almost entirely microspore (fig. 4 *E*).

In every instance, however, the fungi which developed maximum macrospore production under a filter transmitting shorter waves than 2650 A.u. gave increased sporulation with vitaglass also. Therefore this filter seems to be the most advantageous one to use for this purpose. The tests with *F. semitectum* B. & Rav., which were summarized for table III, show no increase in macrospore percentage under vitaglass, but in most individual experiments there was some increase, as is shown by the average in table I.

In the tests with *F. bulbigenum* another result is shown in the percentages cited in both table III and figure 5 for exposure directly to the arc without filters. It will be noted that the average percentage for direct exposure is slightly greater than that for Corex 980 A, making an irregularity in the curve. This irregularity was consistent for all individual experiments with *F. bulbigenum*, as well as for the averages, and was no doubt due to the lethal effects of

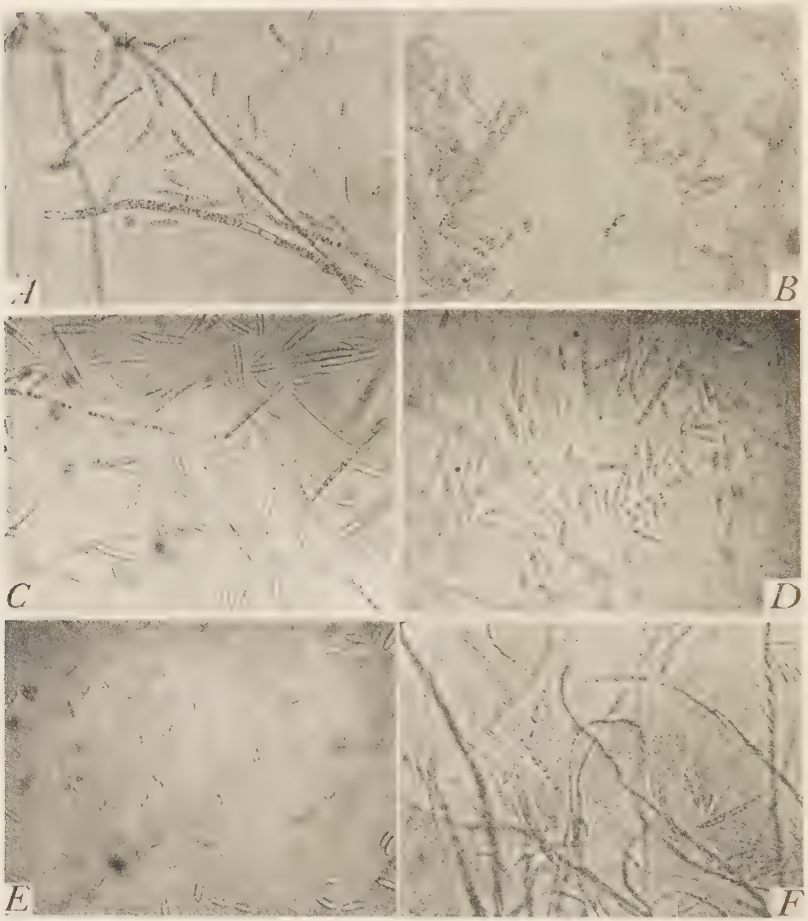


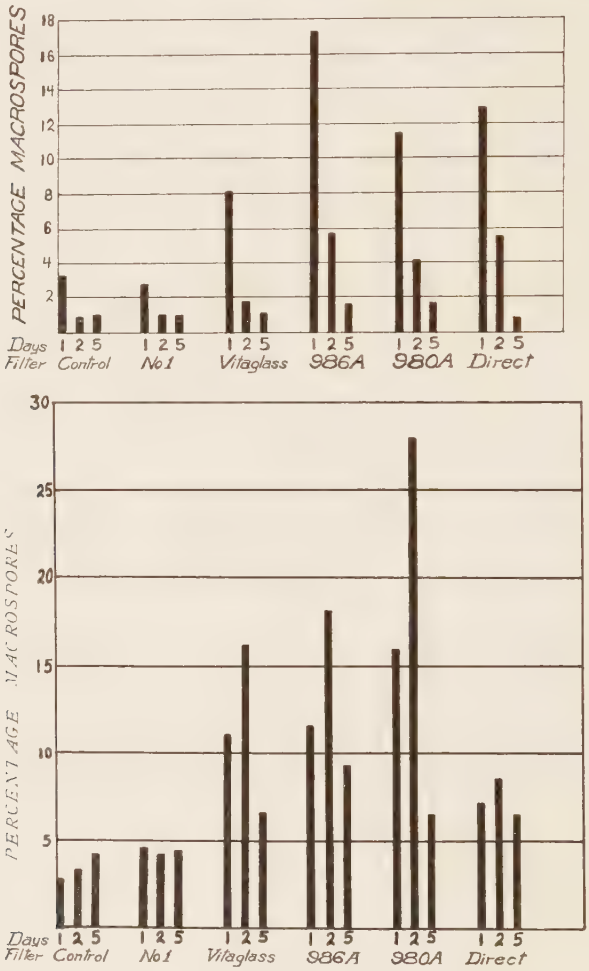
FIG. 4.—Effects of ultraviolet irradiation on cultures of *F. bulbigenum* and *F. sporotrichioides* placed 40 cm. from quartz mercury arc: A, increase in macrospores and decrease in sporotrichiate conidia in culture of *F. sporotrichioides* given three daily 15-minute exposures to Corex red-purple filter 986 A; B, control of *F. sporotrichioides* showing scarcity of macrospores and predominance of sporotrichiate conidia; C, abundance of macrospores of *F. bulbigenum* 24 hours after third daily 15-minute exposure through Corex red-purple filter 986 A; D, spores of *F. bulbigenum* present after three daily 15-minute exposures through vitaglass (2650–6690 A.u.); E, control of *F. bulbigenum* showing absence of macrospores; F, increase in microspore content of *F. bulbigenum* 48 hours after third daily 15-minute exposure through Corex red-purple filter 986 A (2535–4340 A.u.).

the shorter waves transmitted. In all plates exposed directly to the arc, this fungus showed a high percentage of spores whose contents were broken up into globules or which were apparently empty of protoplasm. Mycelium also exhibited a similar condition, but spores seemed to break up more readily and many spores were disrupted or badly shattered. Sometimes it was possible to recognize, for counting purposes, a macrospore which was disrupted or shattered, but shattered microspores could not be so easily detected. Often great masses of loose material or small clumps of floating glycogen and fat particles were suspected of coming from shattered spores, but could not, of course, be interpreted for counting. This decrease in detectable microspores could easily account for the upward trend of macrospore percentage when the plates were exposed directly to the arc.

Such a result occurred also with *F. nivale*, and with *F. semitectum*, which really had its maximum production of macrospores under Corex 980 A but which would seem from the average percentages shown in table III to be most favored by direct exposure. In reality *F. semitectum* was even more subject to destruction by the direct rays than was *F. bulbigenum*. The colony was much stunted and the spores badly shattered, again making a representative count impossible. Under Corex 980 A, on the other hand, the spores were normal.

All of the *Fusarium* species tried were injured somewhat by direct exposure to the arc: decided colony stunting, as measured by radial growth (figs. 7, 9, 10, 13); plasmolysis of protoplasts in mycelium and spores (fig. 2 *F*); empty and abnormally shaped cells were noted. In some cells the protoplasm was present as a single comparatively uniform mass; in others numerous small refractive globules appeared. Microchemical reactions indicated the presence of glycogen and fat globules, some of which stained with Sudan III, others of which were partially saponified by sodium hydroxide. However, the severity of injury varied greatly with the species. *F. argillaceum* was extremely susceptible to injury by the shorter waves, those from the direct arc often causing the mycelium to go over almost entirely into production of chlamydospores, and stunt-

ing occurring even when the shortest waves were screened out by vitaglass. In some species there was little spore shattering under full transmission by the lamp, but spore production was checked so



FIGS. 5–6.—Fig. 5 (above), percentage of macrospores of *F. bulbigenum* present at end of one, two, and five days following irradiation with ultraviolet transmitted directly from quartz mercury arc and through various filters. Fig. 6 (below), percentage of macrospores of *F. vasinfectum* var. *zonatum* present at end of one, two, and five days following irradiation with ultraviolet transmitted directly from quartz mercury arc and through various filters.

that the actual number of spores present was lessened, and sometimes no spores occurred (as in *F. coeruleum*) or they were very rare (as in *F. scirpi* var. *acuminatum*). In other species, as in *F. javanicum* and *F. javanicum* Koord. var. *theobromae* (App. & Strk.) Wr., spores were produced between irradiations but were so shattered and abnormal after direct irradiation as to make representative spore count impossible. In no other case was injury as marked under vitaglass as was usually true in the case of *F. argillaceum* (fig. 1 *F*), although in some forms decrease in growth rate did occur with this filter, as is shown by the figures in table III expressing radial increase in millimeters during the three days following first exposures and by figure 8. Although decrease in growth rate sometimes occurred with this filter and often occurred with the two filters transmitting shorter waves (figs. 9, 10), only in rare cases was there any injury to spores and mycelium such as is described here for direct exposure.

In a number of species the total sporulation was somewhat augmented when irradiations were made through filter no. 1, which transmitted to 3020 A.u., and in some the percentage of macrospores was greater. The macrospore percentage of *F. neoceras* was tripled with filter no. 1 and that of *F. vasinfectum* var. *zonatum* sometimes doubled, but for the most part slight results were obtained with filters transmitting no lower than 3020 A.u. Increasing the length of exposure with this filter gave greater macrospore production with *Fusarium* spp., thus corroborating the findings of Ramsey and Bailey (**14**) for *F. cepae* and *Macrosporium*. The total sporulation produced under the three filters transmitting lower than 3020 A.u. was almost invariably greater than on unirradiated plates. Experiments with the Corex D filter in the Stewart health-lamp (wave lengths transmitted given under "Methods") gave essentially the same results as vitaglass. The following percentages of macrospores were obtained for the species named, in irradiated and non-irradiated plates respectively: *F. sporotrichioides*, given three daily 15-minute exposures, — macrospores 56.3%, control 36.5%; *F. semitectum*, exposed, — macrospores 6.5%, control 3.5%; *F. moniliforme* Sheld. var. *subglutinans* Wr. & Rg., exposed, — macrospores 7.2%, controls 4.6%; *F. orthoceras*, exposed, macrospores 1.2%, control (no macrospores); *F. vasinfectum* var. *zonatum*, exposed, — macro-

spores 46.7%, control 5%; and *F. argillaceum*, exposed, — macrospores 23%, control 22%.

In some species, as in *F. culmorum* previously mentioned, the results of irradiation were evident a week after irradiation, but in many species the effect was lost soon after the experiments were concluded. The germination of macrospores resulting in the production of many microconidia accounts for most of this change. In *F. bulbigenum* the drop in macrospore percentage for this reason became pronounced on the second day after irradiation ceased (figs. 4 *F*, 5), and by the fifth day all plates were essentially reduced to the same macrospore percentage as in the control plates. Consequently it was imperative for accurate comparison that all spore counts be made as near as possible to the end of the 24-hour period following irradiation.

In some of the other species the maximum production of macrospores was evident on the second day following irradiation, as in the case of *F. vasinfectum* var. *zonatum* (fig. 6). On the third day the percentage began to drop, and by the fifth day the percentages were nearing those of the controls. Usually the rise in macrospore count on the directly irradiated plates of *F. vasinfectum* var. *zonatum* was especially noticeable, although in the experiments averaged in figure 6 the increase is not striking.

Effect of ultraviolet on Fusarium species with proven perfect stage

Some tests were made with those species of *Fusarium* with proven perfect stages, to see whether irradiation with ultraviolet would induce perithecial formation. The results of these experiments are summarized in table IV and need little discussion. The 4-day old cultures were treated as in the previous tests with three 15-minute exposures at 40 cm. from the mercury arc, the rays passing through the vitaglass filter. Essentially the same results were obtained whether the filter was in the lamp or used as a petri-dish cover. A duplicate set of tests was run at the same time, however, in which the cultures were exposed to the direct rays from the mercury arc for 4 seconds at a distance of 21 cm. from the arc. This latter set was run since STEVENS (20) obtained many perithecia of *Glomerella*

cingulata (Stoneman) Sp. & Schrk. by exposing them to direct rays for 4 seconds at 21 cm.

While the results on conidia production were essentially as one would expect from previous experiments, no perithecial development was evident at the conclusion of the experiments. A few tests with 1 second and 5 seconds direct and with 15-minute exposures under the various filters were also negative. STEVENS found the rudiments of perithecia of *Glomerella cingulata* 15 hours after irradiation and obtained mature ascospores in 4 days. In the case of *Colletotrichum lagenarium*, although the plexi began to develop 24 hours or less after irradiation and the perithecia 1–10 days later, he found that sometimes the perithecia did not appear for 23–30 days. Therefore the species of *Fusarium* here tested were held for several weeks to two months. Only in the case of a subculture of REINKING'S strain 129 (**33**, p. 168) of *F. javanicum* Koord. var. *theobromae* (App. & Strk.) Wr. did perithecia develop at all, and then only in one experiment. In the work with two strains of this fungus the mature perithecia of the perfect stage, *Hypomyces ipomoeae* (Hals.) Wr., were found seven weeks after irradiation on two plates of one strain which had been thrice irradiated for 15 minutes; and vestigial perithecia were found in one of the second-strain cultures which had been irradiated directly for 4 seconds on each of three successive days. Other plates similarly treated bore no perithecia, and those found on the plates mentioned occurred only on the glass sides of the petri dishes at the edge of the colony, in the new growth since the irradiation; therefore although controls were also negative there was no real evidence that ultraviolet rays were responsible for the occurrence of the few scattered perithecia which appeared in one test.

In another strain of *Hypomyces ipomoeae*, which was producing perithecia in artificial culture, similar tests brought no increase in perithecial production over the control plates which also produced mature perithecia. It may be worthy of note that growing WINELAND'S two strains (**27**) of *F. moniliforme* together under ultraviolet radiation did nothing to restore their erratic tendency to produce the perfect stage, *Gibberella moniliformis* (Sh.) Wineland. These subcultures were not, however, producing the "blue-black globoid

TABLE IV

SUMMARY OF EXPERIMENTS SHOWING EFFECTS OF ULTRAVIOLET ON SPECIES OF FUSARIUM WITH KNOWN PERFECT STAGE

SPECIES OF FUSARIUM	PERFECT STAGE	RESULTS OF IRRADIATION		CONTROL
		4 SEC. DIRECT, 21 CM. FROM ARC	15 MIN. THROUGH VITAGLASS, 40 CM. FROM ARC	
F. decemcellulare Brick. (*R*)* (s. Spicarioides)	Calonectria rigidiuscula (B. & B.) Sacc.	No colony stunting; no perithecia, no macrospores; fewer microspores than control	No colony stunting; no perithecia, no macrospores; more micro. than control	No perithecia; only 0-septate spores
F. nivale (Fr.) Ces. (*D–L*) (s. Arachnites)	Calonectria graminicola (B. & B.) Wr.	No colony stunting; no perithecia; 70.5% macro.; deeper color than control	No colony stunting; no perithecia; 84.8% macro.; deeper color than control	Few spores; no perithecia 35.5% to 60.2% macrospores
F. equiseti (Cda) Sacc. var. bullatum (Sherb.) Wr. (*R*) (s. Gibbosum)	Gibberella intricans Wr.	No stunting; no perithecia even when held several weeks; no macrospores	Same as 4″ direct	No perithecia; no macrospores
F. sambucinum Fuckel (1 strain from *S* and one from potato. Results identical)	Gibberella pulicaris (Fr.) Sacc.	Much stunting; no perithecia even when held 2 months; spores none to rare macrospores	Much stunting; no perithecia in 2 months; spores few to moderately plentiful; 100% macro.	No perithecia in 2 months; no spores
F. graminearum Schwabe (2 strains *W–L*) (s. Discolor Saubinetii)	Gibberella saubinetii (Mont.) Sacc.	Slight stunting; no perithecia in several weeks; 100% macrospores	No stunting; otherwise as for direct	No perithecia; 100% macrospores
F. lateritium Nees. var. mori Desm. (*W*) (Syn. F. urticearum (Cda) Sacc.) (s. Lateritium)	Gibberella baccata (Wallr.) var. moricola (Ntrs) Wr.	No stunting; total sporulation greater than control; 1.3% macrospores	2% macrospores; otherwise as direct; no perithecia in either	Less than 1% macrospores; no perithecia

* Letters in italics refer to source of culture as given in footnote, table I.

TABLE IV—*Continued*

F. moniliforme Sheldon (Wineland's Y 29) (*J*)	Gibberella moniliformis (Sh.) Winel.	No stunting; no perithecia in 2 months; more spores than control but macrospores rare	No stunting; no perithecia in 2 months; more spores than direct; 1% macrospores	No perithecia in 2 months; macrospores rare
Wineland's Y 15 (*J*)	Gibberella moniliformis (Sh.) Winel.	Slight stunting; no perithecia in 2 months; occasional macrospore; fewer than with vita-glass	Slight stunting; no perithecia in 2 months; less than 1% macrospores	No perithecia in 2 months; macrospores rare
Y 29 & Y 15 on same plate		No perithecia in 2 months	As for direct	No perithecia
F. moniliforme Sheld. var. maius Wr. & Rg. (*R*) (s. Liseola)	Gibberella fujikuroi (Saw.) Wr.	Slight stunting; no perithecia in 2 months; more spores than control; less than 1% macrospores	As for direct except thicker pionnotes and 2.1% macrospores	No perithecia in 2 months; macrospores rare
		At conclusion of irradiation period		
F. javanicum Koord. var. theobromae (App. & Strk.) Wr. (*R*)	Hypomyces ipomoeae (Hals) Wr.	No stunting; no macrospores; no perithecia; fewer micro. than control	Slight stunting; few macrospores; no perithecia	No macrospores; no perithecia
		7 weeks after irradiation period		
		A few macrospores; no perithecia	A few macrospores; perithecia at edge of colony on side of petri dish only	A few macrospores; no perithecia
F. javanicum Koord. var. theobromae (App. & Strk.) Wr. (*R*) 2nd strain from (*R*) labeled	"Hypomyces ipomoeae" but bearing no perithecia on receipt	*At conclusion of irradiation period* Slight stunting; no macrospores; no perithecia	Slight stunting; macrospores less than 1%; no perithecia	No macrospores; no perithecia

TABLE IV—*Continued*

Species of Fusarium	Perfect stage	Results of irradiation		Control
		4 sec. direct, 21 cm. from arc	15 min. through vitaglass, 40 cm. from arc	
		7 weeks after irradiation period		
		A few macrospores and a few vestigial perithecia on edge of petri dish only	A few macrospores; no perithecia	
F. javanicum Koord. var. radicicola Wr. (*R*)	Hypomyces ipomoeae (Hals.) Wr. f. 1 Wr.	No stunting; 2.1% macrospores; no perithecia even after 2 months	No stunting; 5.3% macrospores; no perithecia in 2 months	Less than 1% macrospores; fewer micro. than on exposed plates; no perithecia
F. solani (Mart. pr. p) App. & Wr. var. eumartii (Carp.) Wr. (*S*)	Hypomyces haematococcus (B. & Br.) Wr.	Some stunting; microspores only; no perithecia in 2 months	Slight stunting; microspores only; no perithecia in 2 months	Microspores only; no perithecia in 2 months
F. solani App. & Wr. var. minus Wr. (*R*)	Hypomyces haematococcus var. breviconus Wr.	No stunting; no macrospores; no perithecia in 2 months	No stunting; less than 1% macrospores; no perithecia in 2 months	Microspores only; no perithecia in 2 months
F. solani App. & Wr. var. striatum (Sherb.) Wr. (s. Martiella)	Hypomyces haematococcus var. cancri (Rutg.) Wr.	No stunting; only microspores; no perithecia in 2 months	As for direct	Microspores only; no perithecia in 2 months
F. argillaceum (Fr.) Sacc. (*W*) (s. Ventricosum)	Hypomyces solani Rke. & Berth.	Colony very stunted; macrospores none to rare; no perithecia in 3 months; many chlamydospores	Colony stunted; spores few but mostly macrospores (60–70%); no perithecia in 3 months; many chlamydospores	Spores usually more plentiful in control but 20–40% macrospores; no perithecia in 3 months; chlamydospores rare

bodies" which she found present when they were in good condition for pairing and resultant production of perithecia.

Decrease in growth rate and sporulation

Although there was in practically every instance much colony stunting owing to decrease in growth rate and lethal effects of the shorter rays when the cultures received the full emanations from the lamp, the amount of stunting which occurred with the several filters was variable, as is shown in tables III and IV. The stunting following direct exposure was always accompanied by lethal effects, such as plasmolysis of cells in the mycelium or spore destruction; but only with certain species were such conditions evident upon microscopic examinations of the cultures exposed to rays passing through the filters. The decrease in growth rate, as measured by the radial increase in millimeters during the three days following the first exposure, was usually very slight or lacking when vitaglass was used, and usually greater when the Corex filters 986 A and 980 A were used; but this varied for the species irradiated. It will be noted from table III that *F. nivale* and *F. neoceras* usually showed no stunting except upon direct exposure, but occasionally in the same experiment one plate would show stunting under a given filter and the others not (fig. 12). This was not correlated with variations in percentage of macrospores. Thus not only was there variation among the different organisms in this matter but sometimes the same strain reacted differently. *F. decemcellulare* sometimes showed slightly decreased growth rate under vitaglass, usually not; *F. javanicum* (fig. 11) and *F. javanicum* var. *theobromae* (fig. 7) usually showed some stunting when this filter was used, but sometimes not. *F. vasinfectum* var. *zonatum* usually showed no stunting with vitaglass and often none with the two filters transmitting shorter wave lengths; but sometimes there was slight stunting of the colonies under these last filters (fig. 13). There was always stunting with direct exposure, and usually the radial increase after direct irradiation was begun was largely beneath the surface of the agar, the agar screening out the detrimental shorter waves. This was also true of a great many of the other species. For example, the radial increase during the three days following the first direct exposure of *F. semitectum* was 12 mm.

beneath the surface of the agar and only 3 mm. on the surface, while that of the controls was 15 mm. on the surface. In *F. coeruleum* there was almost no growth above the surface on direct plates (fig. 9; table III). In every experiment normal growth was resumed from sub-agar mycelium or aerial mycelium a day or two after cessation of irradiation.

Although decrease in growth rate and macrospore production were often associated, there were so many exceptions that there seemed to be no definite correlation between the production of macrospores after irradiation and inhibition of growth. Therefore, the results of irradiating many species of *Fusarium* would seem to corroborate the findings of RAMSEY and BAILEY (14) for *Macrosporium* and for *F. cepae*, that increased spore production subsequent to irradiation with ultraviolet is not a direct result of inhibition of growth rate. Surely inhibition of growth rate is not alone responsible, for decreasing the rate of growth by exposure to higher or lower temperatures or by using less favorable media did not produce the same results. Through personal correspondence with Miss STRONG and G. H. COONS, the writer learned that in no case did abundant sporulation accompany the inhibition in growth rate of *Fusarium* species owing to the toxic effect of dyes (2).

HUTCHINSON (6), in his work with the response of *Colletotrichum phomoides* to ultraviolet, found similar lack of correlation between the growth rate and sporulation. With full mercury arc irradiation he always got a retardation of growth. However, this was accompanied by increased sporulation only under exposures of 15 seconds to 2 minutes, whereas sporulation decreased when exposures were for more than 2 minutes. He considered this as an indication that sporulation is an inverse expression of growth rate only within certain limits, and that there is an optimum amount of irradiation causing sporulation. He states that certain monochromatic bands retarded growth and stimulated production of acervuli while with other wave lengths there was initial retardation of growth followed by stimulation and also hastening of sporulation. Certain waves constantly stimulated growth and had no apparent effect upon the time of development of spores; but on the other hand wave lengths

4078 A.u. and 5819 A.u., which produced the most marked stimulation of growth, also caused early development of acervuli. Therefore he concluded that retardation and extreme stimulation of growth are accompanied by early sporulation, while irradiations causing intermediate stimulation of growth have no apparent effect in the development of acervuli.

In the present work with *Fusarium*, some species, such as *F. moniliforme*, *F. bulbigenum* forma 1, etc., which showed a decrease in the size of the colony when irradiated, showed no increase in macrospore production. On the other hand, the radial increases given in table III show that macrospore production was greatly increased when *F. nivale* (fig. 1 *C*), *F. sporotrichioides* (fig. 14), *F. scirpi* var. *acuminatum*, *F. neoceras* (fig. 12), and *F. vasinfectum* var. *zonatum* (fig. 13) were subjected to ultraviolet radiation through vitaglass, even when there was no decrease in the rate of growth during this treatment.

Irrespective of whether inhibition of growth of *F. vasinfectum* var. *zonatum* took place under filters 980 A and 986 A, an increased percentage of normal macrospores always occurred. In contrast, *F. javanicum* var. *theobromae* (fig. 7), which showed much stunting under these two filters, showed very little spore increase; in fact, the vitaglass plates, which showed decrease in growth rate but no microscopic evidence of spore destruction, often showed less total sporulation than the control plates. A culture of *F. sambucinum*, treated for 4 seconds direct on three successive days, grew only 3 mm. during those three days; whereas the controls averaged 13-mm. growth but neither set of plates showed any spores; yet duplicate cultures given three 15-minute exposures to rays no shorter than 2650 A.u. likewise grew only 3 mm. and showed considerable spore production. *F. equiseti* var. *bullatum* sometimes exhibited decreased growth after 15-minute irradiation periods, sometimes not; but in neither case did irradiation increase macrospore production. And in the one experiment where there was considerable decrease in colony size, the control plates averaging 48 mm. diameter at the conclusion of the experiment and the plates that received 15-minute irradiation with waves of 2650 A.u. and above averaging 28 mm.

diameter, the control plates showed a greater total spore production than the exposed ones, and rare macrospores as against no macrospores on the treated plates.

These data and the fact that the acme of spore production, except in a few species, occurs with the vitaglass filter which gives no detectable decrease or, at least, the smallest decrease in growth rate (as measured by radial increase of the colony and by observation of amount of aerial mycelium present), would seem to indicate that the increase in macrospores was the result of stimulation effected by ultraviolet rays rather than an indirect result of the inhibition in growth. Further apparent evidence for this conclusion lies in the fact that if a fungus were one responsive to treatment with ultraviolet, old petri-dish cultures of it, which had completely filled the plate and so were arrested in growth, could be made by irradiation to produce spores even on the old mycelium.

What changes take place in the mycelium subsequent to irradiation and immediately preceding sporulation have not yet been worked out. Tests for change in pH of mycelium or of medium as a result of irradiation were negative. No tests were made for the presence of hydrogen peroxide, although it is known that some workers consider the lethal action of ultraviolet to be an indirect one due to development of hydrogen peroxide in the nutrient substrate exposed to the rays. COONS (1) found that he could use a dilute solution of hydrogen peroxide to replace the light stimulus in the production of fruit bodies of *Plenodomus fuscomaculans*. He states that the action of light was modified and controlled by the condition of the mycelium, which is a resultant of all the acting environmental factors. Macroscopically undetectable differences in mycelium condition may also account for the variation in reaction of *Fusarium* species to ultraviolet, and it is thought that microchemical studies of the mycelium previous to and subsequent to irradiation of responsive species may throw considerable light on this question.

No work was done with intensity of the ultraviolet rays involved, since the object of this study was primarily to determine the usefulness of ultraviolet radiation in taxonomic studies of *Fusarium*. RAMSEY and BAILEY had found in the work on *F. cepae* and *Macrosporium* that, while intensity was an important factor, it apparently

is not primarily responsible for the variation between the different filters in amount of increase in sporulation. SWANN and DEL ROSARIO (26), working with *Euglena*, found that the predominance of the lethal effect of one monochromatic ultraviolet line over that of another given line "is not explicable entirely as a matter of absorption, but involves, in addition, other characteristics of the wave lengths concerned."

Discussion

While the chief value of ultraviolet irradiation of *Fusarium* possibly lies in inducing the production of macrospores of certain species when the macroconidia are lacking in the strains under observation, there is also some promise that ultraviolet radiation may be useful for further verification of systematic identification of some species. The consistency of response to ultraviolet of certain forms which characteristically produce macrospores or fail to produce macrospores upon irradiation seems of some diagnostic value. It is interesting to note in this respect that of the ten strains of *F. moniliforme* and three authentic varieties exposed to ultraviolet treatment, none gave noticeable increase in percentage of macrospores; while *F. neoceras* (33), which is the outstanding species morphologically in section Liseola, also stands definitely apart from the other forms of the same section in its response to ultraviolet by abundant macrospore production. *F. argillaceum* also, which stands alone as the only species in its section, is the only species tested which was greatly injured by the rays passing through vitaglass and which showed the striking tendency to go over into chlamydospore production as a result of irradiation. The contrast in response between vascular parasites and other species in section Elegans has already been discussed.

Variation in reaction to the various filters is scarcely sufficient to be of help in separating species, but the responsiveness or nonresponsiveness to macrospore production and the variation in colors intensified by exposure to ultraviolet might easily be of some help in classification. In colored *Fusarium* species, such as *F. bulbigenum* (fig. 8) and *F. coeruleum* (fig. 9), the colors became increasingly intense with filters transmitting lower wave lengths. In the case of some species whose mycelium was white in culture tubes, colors

developed under irradiation by the shorter rays. The mycelium of *F. javanicum*, for instance, which was white under filters transmitting no lower than vitaglass (fig. 11), became pale violet grey to pale glaucous blue above a white to pearl grey background when irradiated through filter 986 A, deepened to deep glaucous grey under filter 980 A, and was dusky green blue (2) to green blue slate when irradiated direct (fig. 10). It therefore stood out in marked contrast to its variety, *F. javanicum* var. *theobromae*, which remained white even when irradiated directly (fig. 7).

It is felt that while the use of response to ultraviolet radiation as an aid in differentiation of *Fusarium* species is of less value than morphological and pathogenicity studies, it may add one more physiological test in instances of difficult diagnosis. The value of irradiation for inducing macrospore production in short time in many species is beyond question. It must be realized, however, that it cannot always be applied, for there are many cases in which it fails to induce macrospore production.

Summary

1. In most of the 59 species, varieties, and forms of *Fusarium* exposed to the ultraviolet rays transmitted by a Cooper-Hewitt quartz mercury-arc lamp through filters transmitting waves as low as 2650 but no lower than 2300 A.u., there was an increase in total sporulation (macrospores and microspores) if not in the percentage of macrospores present. This increase was usually evidenced by the production of a pionnotes or of sporodochia; or at least by greater intensity of mycelium, stroma, and spore mass colors. Colors not present in the control developed in the mycelium of some forms when exposed to the shorter rays of ultraviolet.

2. In some species there was a slight increase in spore production with filters transmitting no lower than 3020 A.u., but with filters transmitting no ultraviolet results were negative.

3. Some species, such as *F. neoceras*, *F. cepae*, *F. solani* var. *medium*, *F. sporotrichioides*, *F. vasinfectum* var. *zonatum* and forma 1 and 2, *F. bulbigenum*, and *F. redolens*, always gave very striking increase in macrospore percentage with three daily 15-minute treatments under vitaglass (transmitting to 2650 A.u.).

4. Some other species gave favorable although not such striking results in macrospore increase.

5. Many species, such as *F. conglutinans*, *F. bulbigenum* forma 1, *F. moniliforme* and its varieties, *F. orthoceras* and its varieties, *F. oxysporum*, etc., showed very little or no macrospore increase after irradiation.

6. Except in the cases of *F. scirpi* var. *acuminatum*, *F. javanicum*, and *F. argillaceum*, the tendency to respond or not to respond to irradiation with macrospore production was consistent and seemed characteristic of the species or variety.

7. All of the species which responded to the treatment, except *F. oxysporum* var. *nicotianae*, were saprophytes or decay producers. The culture of *F. oxysporum* var. *nicotianae* was one which had not been tested for pathogenicity.

8. All of the known vascular parasites which were tested failed to show appreciable macrospore increase as a result of irradiation.

9. The most extensive spore production induced by irradiation occurred in species of sections Sporotrichiella, Gibbosum, Discolor subsection Saubinetii, Martiella, and Elegans subsection Oxysporum (excluding vascular parasite group).

10. Initiation of sporulation in *F. culmorum* was hastened. Irradiated plates showed as high as 83 per cent macrospores whereas controls showed no conidia, and even after the controls began to sporulate, treated plates showed a higher percentage of macrospores.

11. A strain of *F. coeruleum*, which had never produced conidia during its period of culture in this laboratory, and one of *F. sambucinum*, which was temporarily in "abkultur," were induced to produce macrospores after irradiation, although the controls were still sterile.

12. One strain of *Fusarium* (section Gibbosum), isolated from decayed onion bulbs, which had never produced spores in the laboratory, produced many spores (75 per cent macrospores) when irradiated. Subsequent irradiation failed further to increase this percentage.

13. There was little or no cumulative increase in macrospore percentage due to repeated irradiation of non-responsive species, although the quantity of spores present could be thus increased.

14. *F. vasinfectum* var. *zonatum* and *F. semitectum* gave maximum macrospore production under Corex filter 980 A transmitting to 2300 A.u. With *F. sporotrichioides* and *F. bulbigenum* the peak of macrospore production occurred under Corex 986 A transmitting between 2535 and 4340 A.u.

15. Most species tested gave maximum macrospore production under vitaglass. All responsive species gave good results under vitaglass.

16. A 15-minute irradiation period on each of three successive days was better than one longer period. A shorter period was usually not so effective. In some species increasing the length of period to one half-hour, or multiplying the number of 15-minute exposures was advantageous, in others injurious.

17. A distance of 40 cm. from the arc proved best for the purposes of these tests.

18. Exposure directly to the mercury arc, transmitting to 2230 A.u., resulted in decreased growth rate (as measured by radial increase), colony stunting, and injurious effects to mycelium and spores in all species. The extent of the injurious and lethal effects varied with the fungus. In some forms spore formation was inhibited, in others those produced were killed, or injured or deformed. Plasmolysis of protoplasm and shattering of spores occurred.

19. Most forms tried were injured slightly or not at all by the exposure to waves transmitted through the filters, though decrease in growth rate occurred with some species under some filters.

20. Increased macrospore production did not seem to be directly correlated with inhibition in growth rate.

21. A stimulation of macrospore production resulted upon irradiation of old mycelium in cultures of responsive species, although the mycelium already completely filled the petri dish and was checked in growth.

22. *F. argillaceum* usually produced many chlamydospores as a result of irradiation. Cultures exposed directly to the arc sometimes went over almost entirely to chlamydospore production.

23. In some species, as in *F. vasinfectum* var. *zonatum*, macrospore content was greatest 48 hours after irradiation ceased, and there was a drop in percentage subsequently owing to the production of micro-

spores on germination of the macrospores. In other species, as in *F. bulbigenum*, maximum macrospore percentage occurred 24 hours after the third exposure and a drop in percentage was noticeable the second day.

24. Neither three 15-minute exposures through vitaglass at 40 cm. from the arc nor three (or less) 4-second direct exposures at 21 cm. from the arc succeeded in inducing the production of perithecia of the 14 species with known perfect stages which were treated in these tests. Except for two plates in one test of *F. javanicum* var. *theobromae*, none of the cultures tested produced perithecia. In these two plates a few perithecia were present two months after irradiation in mycelium developed since irradiation.

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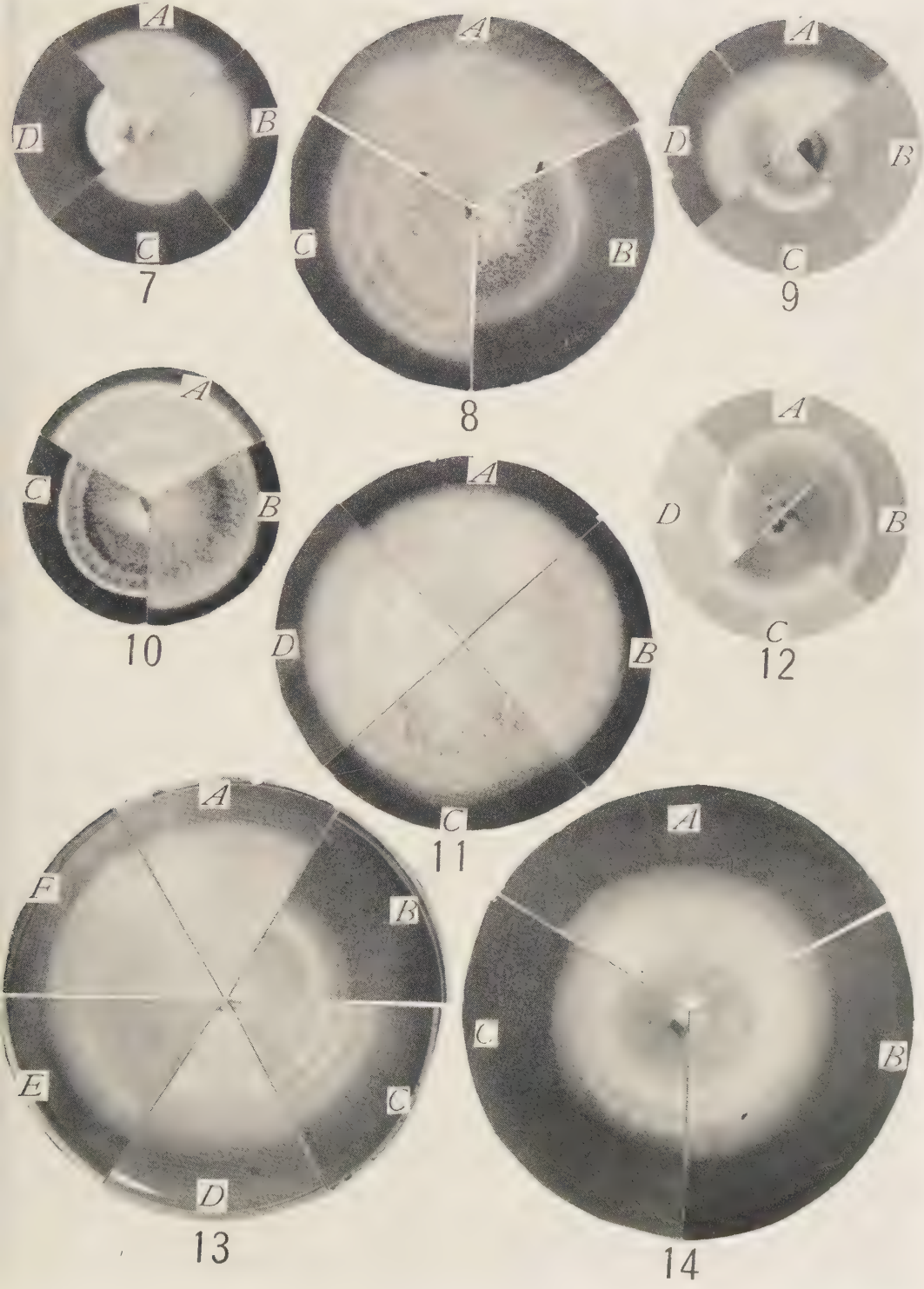
LITERATURE CITED

1. Coons, G. H., Factors involved in the growth and the pycnidium formation of *Plenodomus fuscomaculans*. Jour. Agric. Res. **5**:713–769. 1916.
2. Coons, G. H., and Strong, M. C., The diagnosis of species of *Fusarium* by use of growth-inhibiting substances in the culture medium. Mich. Agr. Exp. Sta. Tech. Bull. 115. 1–78. 1931.
3. Dillon Weston, W. A. R., Studies on the reaction of disease organisms to light. The reactions of disease organisms to certain wave-lengths in the visible and invisible spectrum. Scient. Agr. **12**:352–356. 1932.
4. Dillon Weston, W. A. R., and Halnan, E. T., The fungicidal action of ultra-violet radiation. Phytopath. **20**:959–965. 1930.
5. Fulton, H. R., and Coblentz, W. W., The fungicidal action of ultra-violet radiation. Jour. Agric. Res. **38**:159–168. 1929.
6. Hutchinson, A. H., and Ashton, M. R., The effect of radiant energy on growth and sporulation in *Colletotrichum phomoides*. Can. Jour. Res. **3**:187–199. 1930.
7. Johnson, F. H., Effects of electromagnetic waves on fungi. Phytopath. **22**:277–300. 1932.
8. Link, G. K. K., and Bailey, Alice Allen, Fusaria causing bulb rot of onions. Jour. Agric. Res. **33**:929–952. 1926.

9. NADSON, G., and PHILIPPOV, G., Über die Reizwirkung ultravioletter Strahlen auf das Wachstum von Hefen und Pilzen. Vestnik. rentgenologii i radiologii **5**:425–431. 1927.
10. ———, Action excitante des rayons ultraviolets sur le developpement des levures et des moisissures. Compt. Rend. Soc. Biol. **98**:366–368. 1928.
11. PORTER, C. L., and BOCKSTAHLER, H. W., Concerning the reaction of certain fungi to various wave lengths of light. Proc. Ind. Acad. Sci. **38**:133–135. (1928) 1929.
12. RABINOVITZ-SERENI, D., L'azione dei raggi luminosi visibili di differente lunghezza d'onda sull' accrescimento, sulla sporificazione e sulla pigmentazione dei funghi in coltura pura. Boll. R. Staz. Patol. Veg. Roma n.s. **12**: 81–114. 1932.
13. ———, Il grado di resistenza di alcuni funghi all' azione dei raggi ultravioletti. *Ibid.* **12**:115–144. 1932.
14. RAMSEY, G. B., and BAILEY, ALICE ALLEN, Effects of ultra-violet radiation upon sporulation in *Macrosporium* and *Fusarium*. BOT. GAZ. **89**:113–136. 1930.
15. REINKING, O. A., and WOLLENWEBER, H. W., Tropical Fusaria. Philippine Jour. Sci. **32**:103–253. 1927.
16. RIDGWAY, ROBERT, Color standards and color nomenclature. Published by the author. Washington, D.C. 1912.
17. SIBILIA, CESARE, Due specie di Fusarium parassite di piantine di conifere. Ann. Ist. Sup. Agrar. e Sup. For., Florence. 2d. S. **1**:77. 1924–25.
18. ———, Azione dei raggi ultra-violetti e di alcuni anticrittogamici sui conidii di *Fusarium*. *Ibid.* **1**:145–154. 1925.
19. SHERBAKOFF, C. D., Fusaria of potatoes. Cornell Agr. Exp. St. Mem. **6**: 89–270. 1915.
20. STEVENS, F. L., Effects of ultra-violet radiation on various fungi. BOT. GAZ. **86**:210–225. 1928.
21. ———, The effects of ultraviolet irradiation on various Ascomycetes, Sphaeropsidales and Hyphomycetes. Zentralbl. Bakt., etc. Pt. II. **82**:161–174. 1930.
22. ———, The response to ultra-violet irradiation shown by various races of *Glomerella cingulata*. Amer. Jour. Bot. **17**:870–881. 1930.
23. ———, Relations of nutrients to perithecial production under ultra-violet irradiation. Philippine Agriculturist **19**:265–272. 1930.
24. ———, Further observations regarding ultra-violet irradiation and perithecial development. *Ibid.* **19**:491–499. 1931.
25. ———, The ascigerous stage of *Colletotrichum lagenarium* induced by ultra-violet irradiation. Mycologia **23**:134–139. 1931.
26. SWANN, W. F. G., and DEL ROSARIO, C., The effect of certain monochromatic ultra violet radiations on *Euglena* cells. Jour. Franklin Inst. **213**:549–560. 1932.



BAILEY on FUSARIUM

27. Wineland, Grace O., An ascigerous stage and synonomy for *Fusarium moniliforme*. Jour. Agric. Res. **28**:909–922. 1924.
28. Wollenweber, H. W., Studies on the *Fusarium* problem. Phytopath. **3**:24–50. 1913.
29. ———, Fusaria autographice delineata. Ann. Mycol. **15**:1–56. 1917; Nr. 1–509 ed. 1. 1916.
30. ———, Supplementum Nr. 510–659 ed. 1. 1924; Nr. 1–659 ed. 2. 1926; Nr. 660–1100 ed. 1. 1930.
31. ———, Conspectus Analyticus Fusariorum. Ber. Deutsch. Bot. Ges. **35**: 732–742. 1918.
32. ———, Fusarium-Monographie. Fungi parasitici et saprophytici. Zeitschr. Parasitenkunde **3**:269–516. 1931.
33. Wollenweber, H. W., and Reinking, O. A., Aliquot Fusaria Tropicalia nova vel revisa. Phytopath. **15**:155–169. 1925.
34. Wollenweber, H. W., Sherbakoff, C. D., Reinking, O. A., Johann, Helen, and Bailey, Alice A., Fundamentals for taxonomic studies of *Fusarium*. Jour. Agric. Res. **30**:833–843. 1925.

EXPLANATION OF PLATE I

Segments of petri-dish cultures of *Fusarium* spp. showing comparative growth rates as affected by three daily 15-minute exposures to ultraviolet rays transmitted by quartz mercury arc through various filters (distance 40 cm.).

Fig. 7.—*F. javanicum* var. *theobromae:* A, control grown in diffuse light; B, exposed through vitaglass; C, exposed through Corex red-purple filter (986 A); D, exposed direct.

Fig. 8.—*F. bulbigenum:* A, control; B, exposed through filter 986 A; C, exposed through vitaglass.

Fig. 9.—*F. coeruleum:* A, control; B, exposed direct; C, exposed through Corning Corex filter (980 A); D, exposed through vitaglass.

Fig. 10.—*F. javanicum:* A, exposed through filter 986 A; B, exposed through filter 980 A; C, exposed direct.

Fig. 11.—*F. javanicum:* A, control; B, exposed through filter No. 1; C, exposed through vitaglass; D, exposed through Noviol C.

Fig. 12.—*F. neoceras:* A, control; B, exposed through vitaglass (no stunting); C, exposed through filter 986 A and showing no stunting; D, exposed through filter 986 A but showing stunting. Both plates sporulated equally well.

Fig. 13.—*F. vasinfectum* var. *zonatum:* A, control; B, exposed direct; C, exposed through filter 980 A; D, exposed through filter 986 A; E, exposed through vitaglass; F, exposed through filter No. 1.

Fig. 14.—*F. sporotrichioides:* A, control; B, exposed through vitaglass; C, exposed through filter 986 A.